

Digitized by the Internet Archive  
in 2025

[https://archive.org/details/bwb\\_S0-ELG-628](https://archive.org/details/bwb_S0-ELG-628)













MEMOIRS  
OF  
THE HORTICULTURAL SOCIETY  
OF NEW YORK  
VOLUME 3

PAPERS

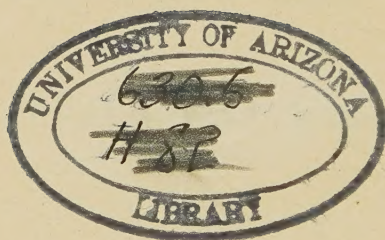
PRESENTED AT THE  
International Conference on  
Flower and Fruit Sterility

August 12-14, 1926



• ISSUED JULY, 1927

COPYRIGHT 1927, BY THE  
HORTICULTURAL SOCIETY OF NEW YORK





P A P E R S

PRESENTED AT THE

INTERNATIONAL CONFERENCE ON  
FLOWER AND FRUIT STERILITY

August 12-14, 1926

PUBLISHED BY

THE HORTICULTURAL SOCIETY OF NEW YORK

NEW YORK

1927

62166

PRESS OF  
LISSO & HARTIG, NEW YORK

Printed in U. S. A.



387, 2  
T61

# THE HORTICULTURAL SOCIETY OF NEW YORK

## INCORPORATED, 1902

*Offices: Bankers Trust Bldg., 598 Madison Ave., New York, N. Y.*

### President

MR. T. A. HAVEMEYER, New York City

### Vice-Presidents

DR. N. L. BRITTON

MR. J. W. CROMWELL

DR. E. B. SOUTHWICK

### Honorary Vice-Presidents

MR. E. S. HARKNESS

MR. F. E. DIXON

MRS. CHARLES H. SENFF

MR. ALBERT C. BURRAGE

MRS. F. A. CONSTABLE

MRS. PAYNE WHITNEY

MR. DANIEL GUGGENHEIM

MR. PIERRE S. DUPONT

MR. W. R. COE

COL. W. B. THOMPSON

### Secretary-Treasurer

MR. F. R. NEWBOLD, 598 Madison Ave., New York

### Assistant Treasurer

MR. EDWARD A. LEROY, JR.

### Executive Secretary

MRS. ELIZABETH PETERSON

### Board of Directors

MR. F. R. PIERSON, *Chairman*

DR. N. L. BRITTON

MR. J. W. CROMWELL

MR. R. T. BROWN

MR. T. A. HAVEMEYER

MR. W. A. DELANO

MR. I. S. HENDRICKSON

MR. F. R. NEWBOLD

COL. W. B. THOMPSON

MR. J. E. LAGER

MR. JOHN SCHEEPERS

MRS. MORTIMER J. FOX

MR. E. A. LE ROY, JR.

MRS. SAMUEL SLOAN

MRS. WM. A. LOCKWOOD

MR. J. A. MANDA

MR. RICHARDSON WRIGHT

MRS. HAROLD I. PRATT

MR. JAMES STUART

MR. F. L. ATKINS

DR. E. B. SOUTHWICK

### Members of the Women's Auxiliary

MRS. HUGH D. AUCHINCLOSS

MRS. E. S. HARKNESS

MRS. C. LEDYARD BLAIR

MISS ANNE MORGAN

MRS. GUY FAIRFAX CARY

MRS. HAROLD I. PRATT

MRS. L. S. CHANLER

MRS. H. L. SATTERLEE

MRS. W. K. DRAPER

MRS. C. H. SENFF

MRS. PAUL FITZSIMONS

MRS. C. H. STOUT

MISS EDITH WETMORE

COMMITTEES FOR THE INTERNATIONAL CONFERENCE  
ON FLOWER AND FRUIT STERILITY

Executive Committee

DR. N. L. BRITTON, *Chairman*

MR. F. R. NEWBOLD, *Treasurer*

DR. A. B. STOUT, *Secretary*

Local Advisory Committee

PROFESSOR R. A. HARPER

DR. C. STUART GAGER

PROFESSOR H. M. RICHARDS

DR. WILLIAM CROCKER

MR. LEONARD BARRON

Entertainment Committee

MR. JOHN SCHEEPERS, *Chairman*

MRS. SAMUEL SLOAN

MRS. HAROLD I. PRATT

MR. T. A. HAVEMEYER

MR. W. R. COE



## PREFACE

The papers herewith published were presented or read by title at the International Conference on Flower and Fruit Sterility which was held under the auspices of The Horticultural Society of New York on August 12th to 14th, 1926. A full report of this Conference as to its organization, complete program, and registered attendance has been printed in the Year Book of The Horticultural Society for 1926-1927. Many of the subjects of the program were necessarily presented to the Conference as summaries of the more complete papers here published. Several of the papers presented or read by title have for various reasons not been submitted for publication by choice of the respective authors.

The editor is pleased to acknowledge the assistance of Mrs. Elizabeth Peterson and Miss Josephine Hauge in the preparation of this volume.

A. B. STOUT,  
*Editor.*



## CONTENTS

	PAGE
BRITTON, N. L. Opening Address .....	1
HAVEMEYER, T. A. Address of welcome .....	2
STOUT, A. B. Types of sterility in plants and their significance in horti- culture .....	3
TISCHLER, GEORG. Investigations concerning the causation of gametic sterility .....	9
LONGLEY, A. E. Relationship of polyploidy to pollen sterility in the genera <i>Rubus</i> and <i>Fragaria</i> .....	15
SWINGLE, WALTER T. Seed production in sterile <i>Citrus</i> hybrids—its scien- tific explanation and practical significance .....	19
HARRISON, J. W. HESLOP AND BLACKBURN, KATHLEEN B. The course of pollen formation in certain roses, with some deductions therefrom..	23
CROCKER, WILLIAM. Dormancy in hybrid seeds .....	33
NORTON, J. B. S. Sterilities and seed production in dahlias.....	39
BEAL, A. C. The role of sterilities in the breeding of floricultural plants..	41
SAUNDERS, A. P. Sterilities encountered in the breeding of peonies....	45
CONNORS, C. H. Sterilities in carnations with special reference to inter- sexes (with plate 1) .....	51
NICOLAS, J. H. Sterility encountered in rose breeding.....	55
REED, ERNEST: Sterility and inbreeding in beets .....	59
MURNEEK, A. E. Correlation and cyclic sterility in <i>Cleome</i> .....	65
ARTHUR, JOHN M. and GUTHRIE, JOHN D. Effect of light, carbon diox- ide and temperature on flower and fruit production (with plates 2-3)..	73
VINALL, H. N. Partial sterility in hybrids of sorghum and Johnson grass (with plate 4) .....	75    ×
CHITTENDEN, FRED J. Sterility in fruits: A summary of twenty years of study at the Royal Horticultural Society's Gardens.....	79
FLORIN, RUDOLF. Pollen production and incompatibilities in apples and pears (with plates 5-7).....	87
CRANE, M. B. Studies in relation to sterility in plums, cherries, apples and raspberries (with plates 8-12).....	119
HEINICKE, A. J. Some factors to be considered in the practical applica- tion of sterility studies of fruits.....	135
MACDANIELS, L. H. An evaluation of certain methods used in the study of the pollination requirements of orchard fruits .....	139    ×
OVERHOLSER, E. L. Apple pollination studies in California .....	151
WELLINGTON, RICHARD. The results of cross-pollination between differ- ent varieties of apples, plums, pears and cherries .....	165



TUFTS, W. P., HENDRICKSON, A. H. and PHILP, G. L. Field studies of the pollination requirements of certain deciduous fruits under California conditions .....	171
PASHKEVITCH, W. Studies on the sterility of the fruit trees in Russia...	175
DARROW, GEORGE M. Sterility in the strawberry and its solution.....	191
LLOYD, FRANCIS E. Abscission in general and with special reference to the curtailment of fruitage in <i>Gossypium</i> (with plate 13).....	195
SCHUSTER, C. E. Sterility in filberts .....	209
SWINGLE, WALTER T. Vegetative and fruiting branches in the date palm and sterile intermediates between them .....	213
CONNORS, C. H. Sterility in peaches .....	215
SUSA, TORASABURO. Sterility in certain grapes (with plate 14).....	223
HANSEN, N. E. Some sterile and fertile plant hybrids .....	229
KIKUCHI, AKIO. Self and cross-sterility in the Japanese pear.....	233
KUNKEL, L. O. Sterility caused by the Aster Yellows Disease (with plate 15) .....	243
BUCHHOLZ, J. T. and BLAKESLEE, A. F. Pollen-tube behavior with reference to sterility in <i>Datura</i> (with plates 16-18) .....	245
MYERS, C. E. The role of sterility in the improvement of vegetables (with plate 19) .....	261
WORK, PAUL. Nutritional factors in seed and fruit formation in vegetable crop plants .....	267
THOMPSON, H. C. Environmental factors affecting seed-stalk development in celery (with plates 20-21) .....	273
DETJEN, L. R. Sterility in the common cabbage ( <i>Brassica oleracea</i> L.)...	277
KOTOWSKI, FELIKS. Effect of self-fertilization in cabbage and onion...	281
PIETERS, A. J. Self-sterility or fertility in <i>Trifolium</i> and <i>Melilotus</i> ....	285
CLARK, CHARLES F. Types of sterility in wild and cultivated potatoes...	289
ARTSCHWAGER, ERNST. Micro and macrosporogenesis in sugar beet with special reference to the problem of incompatibility .....	295
JONES, D. F. Manifestations of impotence in a plant propagated by seed (with plates 22-23) .....	299
BLAKESLEE, A. F., and CARTLEDGE, J. L. Sterility of pollen in <i>Datura</i> (with plate 24) .....	305
LEHMANN, ERNST. The heredity of self-sterility in <i>Veronica syriaca</i> (with plates 25-27) .....	313
EAST, E. M. and MANGELSDORF, A. J. The genetics and physiology of self-sterility in <i>Nicotiana</i> .....	321
SIRKS, M. J. The genotypical problems of self- and cross-incompatibility	325
STOUT, A. B. Studies of the inheritance of self- and cross-incompatibility (with plates 28-29) .....	345

# CONTENTS

xi  
PAGE

SHULL, GEORGE H. Inherited pollen-sterility in shepherd's purse (with plate 30) .....	353
BRIEGER, F. G., and MANGELSDORF, A. J. Linkage between morphological characters and factors for self-sterility .....	369
GAGNEPAIN, F. Why <i>Lysimachia Nummularia</i> L., always very floriferous, is ordinarily sterile .....	373
BOIS, D. Concerning the sterility of phanerogamic plants. (French studies) .....	377
KVAALE, ERLING. Abortive and sterile apple pollen .....	399



## OPENING ADDRESS

N. L. BRITTON

*Chairman, Executive Committee of the Conference*

In selecting a topic for an international conference at this time, the Executive Committee appointed by the Council of The Horticultural Society of New York has been impressed with the high importance of the one chosen, from its economic as well as its aesthetic and scientific aspects. The number and the character of the communications to be presented have evidently fully justified this Conference on Flower and Fruit Sterility. That it has been possible to assemble so large and representative a body of investigators is most gratifying to the Executive Committee, and to the Advisory Committee, and on their behalf I tender you all our high appreciation of your valued cooperation.

The papers to be presented will cover nearly the whole field of study and of observation in this relatively new botanical subject, new, at least, in our recognition of its significance. It seems certain that they will notably contribute to the formulation of general principles and lead by discovery or by suggestion to facts of benefit to mankind.

This Conference is the third to be held under the auspices of The Horticultural Society of New York. The subjects discussed at the first one, held in 1902, were "Plant Breeding and Hybridization"; the second, convened in 1907, was devoted to "Plant Hardiness and Acclimatization." The papers read were published as volumes 1 and 2 of the *Memoirs of the Society*; those to be presented at this time will form volume 3.



## ADDRESS OF WELCOME\*

T. A. HAVEMEYER

*President, The Horticultural Society of New York*

It is with deep regret that absence from home prevents me from welcoming you personally in the name of The Horticultural Society of New York to our city, but our welcome is none the less warm and our appreciation of your presence and interest in our convention is deeply felt by us. We feel assured that the knowledge and information you will impart will be of everlasting benefit to the sciences of horticulture and agriculture; two sciences so closely related that it is difficult to draw the dividing line.

Both agriculture and horticulture mean more to the human race than any of the other sciences, and without them mankind would find it difficult to lead a civilized existence. Our food is dependent upon agriculture, and the beauties of this world are more dependent upon horticulture, perhaps, than anything else in the arts and sciences.

The science of plant breeding means much to the human race, and as the world becomes more thickly populated the growth of our foods becomes a greater problem; as our population increases and our farm lands decrease in area it is most important that each acre should produce its maximum amount of grain, vegetables, or fruit. It is therefore necessary that the plant breeder should develop and improve our crops in such a way that the maximum can be produced per acre.

Europe has developed her land by intensive cultivation and fertilization in such a manner that great crops are produced and the day is not far distant when the United States will have to follow her lead if we expect to be self-supporting.

This Conference should be of great benefit to all plant breeders and therefore of great benefit to all as the improvement and development of all flowers, grain, fruit and vegetables will be greatly helped, we feel sure, by the information gained from the eminent scientists here assembled.

In closing we again wish to extend to you the deep appreciation and thanks of The Horticultural Society of New York for your great interest in our Conference. We trust that you will have a most enjoyable and interesting trip, and we are certain that the many papers presented will be of great benefit to horticulture and agriculture.

---

\* Presented to the Conference by Mr. F. R. Newbold.

# TYPES OF STERILITY IN PLANTS AND THEIR SIGNIFICANCE IN HORTICULTURE

A. B. STOUT

*New York Botanical Garden*

When we speak of a *type of sterility* we refer to some well-defined and frequently recurring condition which breaks that chain of processes that leads normally to fruit and seed production.

It seems that man's interest in sterilities is nearly always first attracted to the influence of environment and culture. This is perhaps only natural for the climatic factors rigidly determine the areas of the profitable culture of any crop or garden plant. Also of obvious, direct, and often spectacular influence on various stages of pollination, fertilization, and fruiting are such factors as unfavorable weather, relations of insects, and ravages of fungi. There are also those intricate and vital relations of the soil to the nurture of the plant, a matter recognized by man's long-continued attempts to regulate and increase productivity through the use of fertilizers.

Frequently certain of the unfavorable environmental factors which constantly threaten a plant may become sufficiently regular in action to constitute a type of sterility. Thus in respect to nutrition there may be what has been termed *plethoric sterility*, a response to such influences as an excess of nitrogen-bearing food materials or even merely of water alone. In general this is characterized by excessive vegetative vigor. The reverse of this condition is an *enervative sterility*.

A rather well-defined type of sterility is the *non-flowering* habit which may develop from a variety of environmental conditions. All about us plants, both wild and cultivated, are frequently and sometimes persistently thrown into the non-flowering condition. By the experimental regulation of light, temperature, and food, the flowering of plants may be controlled quite as nature regulates it for every plant—a matter of special application to the forcing of flower crops. It is a general law, well exemplified by the sweet potato, that plants will thrive vegetatively under environmental conditions which do not admit of their blooming. Then plants automatically become fruitless.

A more precise knowledge of the internal responses of the plant to environmental factors and especially to nutrition, and of the complex internal relations of vegetative vigor to reproductive vigor will, we may hope, lead to more effective methods of culture in promoting fertility. Here is indeed a most worthy field for the further efforts of the plant physiologist.

When we turn to sterilities that are somewhat more fundamentally inherent in the plant itself, we note first that when plants bloom adequately the character of their flowers comes into full play in determining fertility or sterility. There are many types of flowers with reference to the relative development and potency of the two sexes. There are many sorts of plants in respect to the kinds and the relative numbers of flowers they bear. Here the terms fertility and fruitfulness are not necessarily synonymous. A staminate date palm is fruitless but it is highly fertile as a male and plays an important role in the fruiting of the pistillate date tree.

Several well-defined types of sterility exist in respect to the potency of the essential organs in flowers. There is the sterility of double flowers involving the transformation of pistils and stamens into sterile petal-like structures or there may be the complete abortion of these organs. This may involve both pistils and stamens, or either alone, in various degrees and intergradations. Such plants are of course eliminated from food crops grown for fruit or seeds. They are rather numerous among floricultural plants as roses, irises, dahlias, chrysanthemums, peonies, etc., all of which may be propagated vegetatively as clonal varieties. In all such cases the sterility presents decided limitations to the plant breeder who must work through seed-breeding.

Perhaps the type of sterility that has received greatest attention is *sterility of hybridity*. The offspring of wide crosses, such as those of the black currant crossed with the gooseberry, are frequently completely impotent both as males and as females. Hybrids may grow vigorously and bloom abundantly but the spores and gametes which they should yield may not be formed. In such extreme cases the two groups of chromosomes derived from the respective parents live and work in harmony in cells throughout the somatic life of the hybrid only to disintegrate during the intricate processes of spore formation. Sterility from hybridity ranges from complete sterility through various grades of partial sterility. It decidedly limits plant breeding and plays an important role in the evolution of plants.

Sterility of *intersexes* is frequently confused with sterility of hybridity. In intersexes the sterility is typically one-sided and it is frequently extended until there is decided abortion of either the male or the female organs. A plant may produce only poor pollen, as do plants of the Brighton grape and the Green Mountain potato, or the stamens may be completely aborted, as in certain pelargoniums, and yet the pistils of such plants may be able to function fully. In intersexes a race of plants or even an entire species may be decidedly male-sterile as in *Plantago lanceolata*. In fact, there seems to be considerable evidence that maleness is gradually being eliminated in certain hermaphrodite plants. Among fruit crops many clonal varieties derived from male-sterile plants, such as the Brighton grape, the J. H. Hale peach and various pistillate strawberries, set good crops of fruit provided there is proper cross-pollination. The fruit grower must learn to provide for this by proper interplanting.

While intersexes do exist entirely apart from hybridity they may, apparently, appear as disturbances in the development of sex in certain hybrids. But the typical sterility of hybridity differs from the one-sided sterility of intersexes in that it is a fundamental disturbance of chromosomes in the nuclear processes of spore formation and affects male and female elements to the same degree.

Abortion of spores and gametes, similar in effect to that in inter-specific hybridity, may also appear through irregular combinations of chromosomes in certain types of polyploidy, some of which are, we may say, a sort of self-hybridity. Various grades and degrees of abortion of spores may follow this intrusion of unusual sets or numbers of chromosomes into the cell mechanism of a plant. In certain cases such plants can only be increased by vegetative propagation or by their own development of apogamy.

When flowers are produced with the one sex or the both sexes potent to the degree characteristic of the species, the individual plant, or the clonal variety, then the requirements for pollination must be satisfied as an important link in fruit and seed production. Here come into influence on the sterility of hermaphrodites, first, the various and often intricate morphological adaptations for pollination particularly by insects, and second, the physiological adaptations expressed by the maturity of the two sexes in the same flower, or in separate flowers on the same plant, at different times, a condition called dichogamy.

In these cases, especially, unfruitfulness from lack of pollination may develop either when the agents for pollination are not present and operating, or when plants are so placed that the necessary pollinations cannot be made. This latter situation develops whenever a clonal variety that requires cross-pollination is grown in solid blocks. At least three conditions may bring this about.

1. The most obvious case is when a pistillate clonal variety is grown without interplanting. This has already been mentioned in considering intersexes.

2. Such advanced types of adaptation as dimorphism and trimorphism mechanically limit pollination to legitimate crossings. This is best seen among some of our floricultural plants as primulas, lythrums, and orchids, but apparently does not exist to any appreciable degree among important food plants.

3. The condition of sterility is also enforced on clonal varieties when dichogamy decidedly limits pollination from flower to flower among plants of the clonal variety. In certain pecans, for example, the pollen is shed after the female flowers are receptive giving dichogamy that is exclusive and seasonal. The most specialized dichogamy known is in avocados where the development is such that, for certain plants and the clonal varieties developed from them, the hermaphrodite flowers are functionally male only in the forenoon and female only in the afternoon, while plants of other clonal varieties are female in the forenoon and male in the afternoon. In such cases as these, the planting of a clonal variety in solid blocks greatly reduces chances for pollination even



when insects may be abundant and hence the crop is automatically thrown into enforced sterility. Proper interplanting then becomes important to fruit production.

We may pass now to sterility that develops because of the very rigid and precise demands for fertilization. In many plants *physiological incompatibilities* render both self- and certain cross-pollinations ineffective in fertilization. The essential organs of the perfect flowers are fully potent in certain relations but not in others. The plants of a clonal variety developed from a fully self-incompatible plant are of course intra-incompatible. We may picture the situation in an orchard block planted exclusively to the Bartlett pear, the Delicious apple, the German Prune plum, or the Napoleon cherry. The trees may bloom profusely and insects may work these flowers assiduously making many thousands of self- and close-pollinations very few or even none of which will result in the setting of fruit.

The interplanting of two self-incompatible and cross-incompatible varieties, such as the Bing and the Napoleon cherries, does not improve this condition. This type of sterility is quite general among various fruit crops and will quite properly receive considerable attention in the program of this Conference.

It should perhaps be stated that the term "self-sterile" was applied by such earlier writers as Charles Darwin to the condition of physiological incompatibility discussed above. This term will doubtless thus be used in various papers presented in this Conference. But the term is also applied to cases of self-fruitfulness which involve other types of sterility such as seasonal dichogamy and male sterility, particularly the latter. Thus the sugar beets, the Pan American pecan, the J. H. Hale peach, and the Bartlett pear are all called self-sterile. This usage is confusing; it merely indicates that these varieties are self-fruitless without distinguishing the condition responsible; it refers to a result rather than to the cause. In the interests of horticulture, botany, and plant breeding it is highly desirable that the different conditions which have been included in "self-sterility" be determined and clearly designated. Self-sterility that involves only a failure in fertilization may well be called self-incompatibility.

The term "fertile" as generally used in botanical and horticultural literature usually means merely that a plant or a clonal variety is able to yield fruit. The term has a special significance in regard to the behavior of hybrids, but it does not imply that a plant is necessarily male fertile, or self-pollinating, or self-compatible.

As ordinarily grown in field culture, numerous clonal varieties cultivated for such products as tubers, fleshy roots, sugar in sap, etc., frequently fail to produce fruits and seeds. This has very generally been considered as evidence that plants which naturally reproduce asexually by such means as tubers, bulbs, fleshy roots, and rhizomes lose, as a direct result, power for seed reproduction through a degeneration of sex organs (plethoric sterility). When the sex organs of such plants are apparently still fully developed it has

been considered that the vigorous growth of the vegetative organs divert and utilize the available food so that seeds are unable to develop (direct correlative sterility).

The recent studies clearly reveal that the sterilities of many of these plants merely involve such types of sterility as intersexes and self-incompatibility. Thus certain of the cultivated clonal varieties of sugar cane, of which seeds were for many years unknown, and the numerous varieties of the Irish potato are only male sterile while the clonal strains of sweet potatoes and various lilies are merely self-incompatible. They all yield fruit and seed abundantly when properly cross-pollinated. They are fruitless in ordinary culture because the growing of the clonal variety in solid blocks automatically reduces or even entirely prevents cross-pollination. Thus we must recognize that a condition of direct correlative sterility is not operating in these plants. The extent to which it does operate and its role in the fruiting of plants, and especially of cultivated plants, need to be determined.

I am not aware of conclusive evidence that any clonal strain of plants has suffered an inherent and permanent degeneration of one or of both of the sex organs as a direct result of a long continued vegetative propagation. The evidence, such as we have, seems to indicate that even the most extreme types and grades of sterility now seen in any clonal variety existed in the original seedling from which that clon was derived. We need further and more precise knowledge regarding the conditions, the causes, and the hereditary factors involved in the origin and development of seedless fruits.

In the case of seedless fruits we have the situation of sterility with fruitfulness. Such plants may be propagated vegetatively as clonal varieties with only a limitation for use in breeding. A considerable list of important fruit plants, pineapple, navel orange, seedless grapes, banana, etc., are in this class. In some of these, pollination is quite as necessary as in the case of fruits with seeds. In others the fruit is simply a vegetative structure and no pollination or fertilization is necessary for its development. Possibly the ultimate and most ideal goal of horticulture is to develop fruits, like the navel orange, which will mature fruit without any pollination. Sterilities from lack of pollination and fertilization are thus eliminated and the problems of fruit growing thereby greatly simplified.

An interesting situation develops in what is known botanically as apogamy. In this, the embryos in the seeds are practically only buds produced internally. This condition automatically limits the efforts of the plant breeder. The plant which is fully apogamous produces seeds in abundance but is inherently cross-sterile to everything.

There is some evidence that apogamy may be induced in certain plants, either temporarily and occasionally under the stimulus of particular pollinations, or permanently through the development of certain nuclear organizations, especially after hybridization or polyploidy. Many perplexing situations confront the plant breeder in reference to induced apogamy and to one-parental inheritance concerning which precise knowledge is much needed.

The matter of inherent and constitutional capacities for productivity remains to be at least mentioned. Horticultural and agricultural experience is continually determining what strains are most productive of flowers and fruit. For clonal varieties, standardized by vegetative propagation, experience often gives very reliable judgment of inherent productivity provided there are no complications through lack of pollination.

But mere experience is less reliable and effective in the case of annuals grown from seed each year with the crop a population of different individuals among which there is chance for repeated promiscuous cross-breeding. Here there is nearly always a chance to improve productivity. To do this there are at least two lines of attack. One is the selection from pedigreed lines of the most productive strains and the elimination of the unproductive. When these qualities have hereditary values they may be eliminated and the total yield of the crop increased through greater average uniformity. Pedigreed seed-breeding is a recognized method of isolating the most highly productive strains. A second method is that of increasing the inherent capacity for productivity by developing vigor through hybridity, heterosis, or polyploidy. Vegetatively vigorous and highly productive plants may thus be produced. It is indeed quite possible that many of our important cultivated plants have been thus developed. Without doubt these means will be utilized under more fully controlled conditions to the decided benefit of agriculture and horticulture.

To develop plants inherently productive and fertile is perhaps the special task of the plant breeder and the geneticist. To provide proper culture for such plants is the special task of all those who grow plants for their products. Meanwhile a wide range of plant forms are in cultivation which exhibit the various types of sterility and fertility and have varied demands for pollination, fertilization, and fruit production.

It is indeed a worthy aim of horticultural and botanical effort to discover as precisely as possible the condition responsible for each case of persistent sterility or unfruitfulness. It is only thus that we may hope to deal adequately with sterilities either in the commercial growing of crops or in the breeding of plants for greater fertility.

The problems demand no less than the utmost knowledge and skill of the plant breeder, the continued scientific efforts of the botanist, and the keen experience and best art of the practical horticulturist.



# INVESTIGATIONS CONCERNING THE CAUSATION OF GAMETIC STERILITY\*

GEORG TISCHLER

*University of Kiel, Germany*

During the past twenty-three years I have busied myself with questions regarding the causes of gametic sterility. I have endeavored to attack these problems from many sides, and shall briefly report upon this work as follows:

The study of hybrids seems to me especially suited as an introduction to these problems. And I found in the totally sterile hybrid *Ribes Gordonianum* (*Ribes aureum*  $\times$  *sanguineum* [1906a]) as well as in the hybrid *Bryonia alba*  $\times$  *dioica* (1906b), raised by Correns, far-reaching disturbances during the reduction division of the pollen mother cells. These are the familiar appearances, which in their time were first made known through the investigations of Juel (1900) for *Syringa chinensis*, such as disturbance in the transportation of chromosomes, formation of extra nuclei, appearance of double spindles, an absence of sufficient plasm, poor development of the spindle figure, etc. Furthermore, in the *Bryonia* hybrid I also called attention to the fact that the nucleoplasmic relation, namely, the normal ratio between the mass of nucleus and the mass of cytoplasm, can no longer be brought about in the young pollen. These factors bring about that the very greatest part of the pollen is "blind." In other words gametic sterility results.

I also followed the fate of the embryo-sac mother cells and in so doing disposed of the older conceptions according to which the macrospores of hybrids were considered more fertile than the microspores (1903b). In particular I showed that *Ribes Gordonianum* and the above-mentioned *Syringa* hybrid have in fact a practically constant complete obliteration of the embryo sac and the same is also found in the *Bryonia* hybrid. I then pointed out that these findings in themselves did not yet justify the conclusion of *hybridity*. For also external factors can bring about altogether similar destruction of spores. For instance insufficient supply of nutrients for the macrospore mother cells of *Cytisus Adami*—later recognized as a periclinal chimaera—brings about complete obliteration of the embryo sac (1903a). At the same time the pollen grains here are normal, and, as was learned later, represent pure *Laburnum vulgaris* cells.

As far as the already mentioned *Syringa* hybrid is concerned, which is sterile at least in middle Europe, I pointed out (1907, 1908) that also in *Syringa persica* and *S. vulgaris* (looked upon as parent of *S. chinensis*)

---

\* The translation here printed was made by Dr. William Marquette.



"blindness" exists in a large percentage of the spores. But, I also found in the development of pollen that for the greatest part the allotypic mitoses progress perfectly normally, and yet subsequently the gametes become sterile. Something similar I also found in a part of the pollen mother cell of *Ribes* and *Bryonia*. Therefore in these instances the cellular disturbance which otherwise is put forth as an explanation in the case of hybrids cannot be throughout the cause of degeneration of the sex cells. Later Miss Borgenstam (1922) expanded my contentions. She recognized that under uniform external conditions the reduction division may proceed entirely normally, and that the abnormalities found by Juel and myself are to be primarily referred to temperature fluctuations to which *Syringa* is exposed in our region during flower development.

In principle I showed the same as early as 1907 and 1908 for a *Potentilla* hybrid (*Potentilla Tabernaemontani*  $\times$  *P. rubens*) which otherwise is almost fertile throughout. It was possible for me here through the early influencing of the archesporangium to bring about plasma deficiency in the pollen mother cells and consequent disturbance in the divisions, especially in formation of the spindle figure, and thus, for a great part, to make the pollen "blind."

Finally, I have also shown that in *Mirabilis Jalapa*  $\times$  *M. tubiflora* (1907, 1908) produced by Correns, the reduction division may be entirely normal and yet gametic sterility finally sets in. Here, however, I observed an unusual occurrence in the early "blinding" pollen, namely, the membrane did not remain thin as usually is the case in degenerating microspores, but rather it developed typically as in fertile pollen with its thick exine differentiated into various layers. I am inclined to assume this takes place under the influence of the well-developed tapetal cells which bound the pollen chamber.

Thereupon, I utilized my preceding experience as well as those of other authors in two more lengthy exposés on the explanation of gametic sterility (1908, 1915). More and more I have decided that misfits in the chromosome sets are responsible for sterility in those cases in which the distribution of the chromosomes is normal, but yet sterility occurs. I pointed out how the cell is doomed to death from the moment there is no longer present in the cell a completely harmonious haploid chromosome set, but only discordant parts of two different sets which cannot complete each other.

In spite of all this, I have never denied that in a large part of all cases the gross morphological disturbances of the reduction divisions already constitute the primary cause of gametic sterility. I discussed this on a broad basis in my "Karyologie" (1921, 1922) and in an article in the "Bibliographia genetica" (1925a). Long before, on the occasion of a study in Java, Ceylon and East Africa, I had opportunity to study carefully a number of *Musa* varieties and described cytologically in detail their very far-reaching disturbances in the reduction divisions of the pollen mother cells (1910b) as well as their final embryo sac obliteration (1912). At the beginning I was

not convinced of the hybrid nature of bananas. But soon, following a suggestion by Winkler (1911), I was led to see in my *Musa* specimens ancient hybrids, which through vegetative propagation are permanently maintained in the  $F_1$  generation. I have shown, however, through occasional normally developed embryo sacs, as well as through the ability to germinate of individual (occasional) pollen grains, that at least occasionally such favorable chromosome combinations may come about in a sex cell that fertilization should be possible. As is well known, D'Angremond (1914) later fully confirmed my conclusions and extended my findings.

For pineapple varieties, which are likewise considered as hybrids in the tropics, I demonstrated (1912) peculiar disturbances in the structure of the ovules, which in their appearance almost suggest tyloses, and which will in part be responsible for an abnormal supply of nutrients and sterility of the egg cells. Haberlandt (1921) much later produced similar formations in *Oenothera Lamarckiana* and called attention to the similarity with my findings in the pineapple.

The fact that gametic sterility can be brought about through influence induced from without, even if before the allotypic mitoses proceeded normally, led me to work with plants which normally form a number of pollen grains that genotypically, in principal, are viable, but none-the-less never can germinate. This pollen remains permanently sterile and seems to serve only as food for the flower visiting insects.

In Buitenzorg, I investigated from this viewpoint special *Cassia* species (1910a, 1917). In *Cassia Fistula* I found the most interesting object. I could easily relieve the sterility of the "fodder anthers" through the addition of saliva or diastase and thus induce the pollen grains, which otherwise never germinate, to germinate. For under the influence of this addition, perhaps as a result of the penetration of ferments, perhaps also after inciting an autogenous production of diastase, the starch grains are dissolved and the proper condition of maturity for sprouting of the pollen tubes is reached. v. Faber (1912) later showed exactly the same for *Psychotria bacteriophila*.

But beside this I had observed in *Cassia Fistula* and in the other *Cassia* species especially in *C. bacillaris*, that precisely the largest and best appearing pollen grains did not germinate, but remained entirely unchanged in the nutrient solutions even when conditions for germination of the smaller grains were met. Exhaustive measurements of the nucleoplasmic relation later showed me (1925b) that precisely in the largest grains the size of the nucleus, or more specifically of the nuclear surface, is "too small" in relation to the mass of cytoplasm. I, therefore, enunciated the hypothesis that the surface of the vegetative nucleus through secretions of enzymes which are capable of mobilizing the yolk material furnishes a determining factor for the germinability of the pollen. Likewise I saw in the frequently occurring amoeboid nuclear forms attempts of the cell to increase the secreting nuclear surface. I attempted to verify this hypothesis by means of experiments with *Primula sinensis* (1925b). While attempting to modify the degree of heterostyly in

this plant (1918), I had noted the ease with which the size of the pollen grains could be influenced. Measurements of the nucleoplasmic relations convinced me even more than in the case of *Cassia* that my above-mentioned reasoning is correct. The decrease in nuclear size with the size of the cell, and the inability of the largest pollen to germinate was striking. The nucleoplasmic relations could shift in this from 1:15 up to 1:200.

Yet something different attracted my attention in *Primula*. In individual grains the vegetative nucleus, that is the "functioning" nucleus for the cell-life, began to divide as we know this occurs for the nucleus of the young microspore. In two succeeding divisions four nuclei were produced. Also de Mol (1923) and other authors have seen the same for "over-nourished" pollen of various species. Indeed Němec (1898) many years ago had once found in *Hyacinthus* that even eight nuclei appeared which thereupon arranged themselves as in the embryo sac. I saw in such cases a tendency towards a revamping of the microspore into a macrospore. In this connection the familiar experiments of Shattuck (1910) with *Marsilia* should also be recalled. There would thus be achieved a reversal of sex in the gametophyte, phenotypically although not genotypically, a possibility for which above all others Schaffner (1925) has successfully contended.

*Primula sinensis* is genotypically uniform in all its pollen grains, whereas in the dioecious plant 50% carry an X-chromosome and 50% a Y-chromosome. A closer examination of *Melandrium album* and *M. rubrum* (1925c) showed me that here also my hypothesis holds. The nuclear portion ranks steadily with the cell size and the largest pollen grains showed a much smaller percentage of germination than the smaller. The viability of the pollen could be greatly affected experimentally. For that purpose I sowed ripe pollen on sugar gelatine and exposed it to alcohol fumes. Correns had shown that if pollen thus treated is applied to stigmas a higher percentage of males arises than is normally the case. Now I found that after alcohol treatment almost exclusively the smaller grains germinate, the larger ones, including the size classes which normally still germinate, were accordingly eliminated from the competition. It is clear that in this manner a difference in the resistance of the pollen to poison coming from without must be obtainable and with it a graduated sterilization. And again I am of the opinion that the decisive element is the size of the vegetative nuclei present in the pollen. The future must decide to what extent the further questions raised by this are to become the point of departure for new knowledge.

#### LITERATURE CITED

- d'Angremond, A., 1914. Parthenokarpie und Samenbildung bei Bananen. *Flora* 107: 57-110.  
 Borgenstam, Elsa, 1922. Zur Zytologie der Gattung *Syringa*. *Arkiv. Bot.* 17: 1-27.  
 Correns, C., 1922. Alkohol und Zahlenverhältnis der Geschlechter bei einer getrenntgeschlechtigen Pflanze (*Melandrium*). *Naturwissenschaften* 10: 1049-1052.  
 Faber, F. C. von, 1912. Das erbliche Zusammenleben von Bakterien und tropischen Pflanzen. *Jahrb. Wiss. Bot.* 51: 283-375.  
 Haberlandt, G., 1921. Ueber experimentelle Erzeugung von Adventivembryonen bei *Oenothera Lamarckiana*. *Sitz. Akad. Wiss. Berlin* 40: 695-725.



- Juel, H. O., 1900. Beiträge zur Kenntniss der Tetradentheilung. *Jahrb. Wiss. Bot.* **35**: 626-659.
- de Mol, W. E., 1923. Duplication of generative nuclei by means of physiological stimuli and its significance. *Genetica* **5**: 225-272.
- Němec, B., 1898. Ueber den Pollen der petaloiden Antheren von *Hyacinthus orientalis* L. *Bull. Int. Acad. Sci. Prague* **5**: 17-23.
- Schaffner, J. H., 1925. Sex determination and sex differentiation in the higher plants. *Am. Nat.* **59**: 115-127.
- Shattuck, Charles H., 1910. The origin of heterospory in *Marsilia*. *Bot. Gaz.* **49**: 19-40.
- Tischler, Georg, 1903 *a*. Ueber eine merkwürdige Wachstumserscheinung in den Samenanlagen von *Cytisus Adami* Poir. *Ber. Deutsch. Bot. Gesell.* **21**: 82-89.
- 1903 *b*. Ueber Embryosack-Obliteration bei Bastardpflanzen. *Beihefte Bot. Centralblatt* **15**: 408-420.
- 1906 *a*. Ueber die Entwicklung des Pollens und der Tapetenzellen bei Ribes-Hybriden. *Jahrb. Wiss. Bot.* **42**: 545-578.
- 1906 *b*. Ueber die Entwicklung der Sexualorgane bei einem sterilen Bryonia-Bastard. *Ber. Deutsch. Bot. Gesell.* **24**: 83-96.
- 1907. Weitere Untersuchungen über Sterilitätsursachen bei Bastardpflanzen. *Ber. Deutsch. Bot. Gesell.* **25**: 376-383.
- 1908. Zellstudien an sterilen Bastardpflanzen. *Arch. Zellforschung* **1**: 33-151.
- 1910 *a*. Untersuchungen über den Stärkegehalt des Pollens tropischer Gewächse. *Jahrb. Wiss. Bot.* **47**: 219-242.
- 1910 *b*. Untersuchungen über die Entwicklung des Bananen-Pollens. I. *Arch. f. Zellforschung* **5**: 622-670.
- 1912. Ueber die Entwicklung der Samenanlagen in Parthenokarpen Angiospermen-Früchten. *Jahrb. Wiss. Bot.* **52**: 1-84.
- 1915. Chromosomenzahl, -Form und -Individualität im Pflanzenreiche. *Progressus Rei Bot.* **5**: 164-284.
- 1917. Pollenbiologische Studien. *Zeitsch. Bot.* **9**: 417-488.
- 1918. Analytische und experimentelle Studien zum Heterostylie—Problem bei *Primula*. *Festschr. zur Feier d. 100 jähr. Besthens der Kgl. landwirtsch. Hochschule Hohenheim.* 254-273.
- 1921-22. Allgemeine Pflanzenkaryologie. Vol. II. *Handbuch Pflanzenanatomie.* Berlin.
- 1925 *a*. Die cytologischen Verhältnisse bei pflanzlichen Bastarden. *Bibl. Genetica* **1**: 39-68.
- 1925 *b*. Studien über die Kernplasmarelation in Pollenkörnern *Jahrb. Wiss. Bot.* **64**: 121-168.
- 1925 *c*. Ein Beitrag zum Verständnis des Certationsproblems bei *Melandrium*. *Planta* **1**: 332-342.
- Winkler, Hans, 1911. (A review of the article by G. Tischler cited here as 1910 *b*). *Zeitsch. Bot.* **3**: 175-178.





# RELATIONSHIP OF POLYPLOIDY TO POLLEN STERILITY IN THE GENERA RUBUS AND FRAGARIA

A. E. LONGLEY

*Bureau of Plant Industry, U. S. Department of Agriculture*

Previously published articles have established two facts concerning the morphological character of pollen in the genus *Rubus*. (1) Many species and forms have a high percentage of poor pollen (1, 2, 3). (2) Variations occur in the chromosome number of many forms (4, 5, 6).

The present investigation of pollen sterility and chromosome number includes over fifty wild species and horticultural varieties.

A relationship has been observed between pollen sterility and irregularities in the reduction phases at the time of pollen formation.

The following table classifies the forms studied, showing their distribution among the various chromosome groups and the average percentage of sterility occurring in these groups:

TABLE 1  
NUMBER OF FORMS AND PERCENTAGE OF STERILITY IN VARIOUS CHROMOSOME GROUPS

		CHROMOSOME GROUP						
		7	21/2	14	35/2	21	49/2	28
Horticultural variety	No. of forms. ....	10	0	26	0	6	0	4
	Average per cent of sterility	13.8	.....	22.7	.....	6.0	.....	3.5
Wild species and forms	No. of forms. ....	4	12	1	2	0	0	0
	Average per cent of sterility	7.6	72.9	20.7	68.6	.....	.....	.....

The first line of the table shows that no form having an odd multiple of the basic chromosome number seven is found among the 46 horticultural varieties. The table also shows that the average amount of pollen sterility in horticultural forms is less than 25 per cent.

The presence of only diploid, tetraploid, hexaploid, and octoploid varieties and the absence of forms with any large amount of pollen sterility among these cultivated *Rubi* seems to suggest that an association exists between even chromosome numbers and the production of fertile pollen. The presence of an even number of chromosomes in pollen mother cells allows an equal distribution of the chromosome elements among the four daughter pollen grains, a behavior probably essential to the production of good pollen.

The association between even chromosome number and the production

of good pollen is also apparent in the five diploid and tetraploid wild species investigated. The table shows that the average pollen sterility in these forms is well below 25 per cent.

Conditions are quite different in forms having  $21/2$  and  $35/2$  chromosomes. In varieties with these odd chromosome numbers it is evident that irregularities must occur in the chromosome distribution to the four daughter cells. One sees also that in triploid and pentaploid forms there is very little fertile pollen. This seems to indicate a relationship between odd numbers of chromosomes and the production of sterile pollen.

The situation is as follows: *Rubus* forms belonging to the even diploid, tetraploid, hexaploid and octoploid groups have very little sterile pollen, while forms having such incompatible chromosome numbers as triploid and pentaploid produce very little good pollen.

The contrasting morphological characters of even and odd chromosomed varieties suggest that the former have originated from the union of sex cells quite similar in character and having the same chromosome numbers. The behavior of the chromosomes in several tetraploid plants seems to verify this assumption. At diakinesis there is usually 7 tetravalent chromosomes instead of 14 bivalents as expected. The presence of 7 instead of 14 homologous chromosomes in a form seems strong evidence that a close relationship existed between the parent forms.

On the other hand the behavior of the chromosomes in forms with odd numbers and with the associated high percentage of sterile pollen suggests that such forms originated by the union of distantly related sex cells having different chromosome numbers.

In *Rubus*, therefore, a large percentage of good pollen in a form indicates a close relationship between parent forms and the presence of an even chromosome number, while a small percentage of good pollen in a form predicts the presence of an odd chromosome number that has originated by the union of cells with different chromosome numbers.

The absence of triploid and pentaploid forms among the horticultural varieties studied finds a natural explanation. The horticulturist as he works for fertile forms, consciously or unconsciously eliminates all with a high percentage of poor pollen and consequently discards all forms belonging to odd chromosomed groups.

In the genus *Fragaria* sterility is not confined to the mere abortion of pollen. In many forms some or all of the pollen bearing organs are reduced to staminodia or are absent, giving in the extreme cases a true dioeciousness.

The author found that all diploid *Fragaria* forms are hermaphroditic and that dioeciousness as it exists in this genus is associated with polyploidy. Lest this statement be misconstrued it is well to point out that the reverse does not hold, for many polyploid species are hermaphroditic.

In order to test further the association between dioeciousness and polyploidy it was suggested that a chromosome determination of the dioecious *Rubus chamaemorus* be made. I was able to procure buds of this species

and found that there are 28 haploid chromosomes in the pollen mother cell. Therefore this dioecious *Rubus* has proved to be hexaploid.

The origin of polyploidy is at present a debated question, but looking upon dioeciousness in *Rubus* and *Fragaria* as a degeneration and a sterilization of either the male or female organs of a hermaphroditic form it seems reasonable to conclude that polyploid forms have arisen from diploid and are consequently of recent origin.

This study of chromosomes in economic forms of *Rubus* has been carried on in cooperation with G. M. Darrow of the Office of Horticulture of the United States Department of Agriculture. We hope that a foreknowledge of the chromosome number in horticultural varieties will be an aid in hybridization.

# BIBLIOGRAPHY

1. Hoar, C. S., 1916. Sterility as the result of hybridization and the condition of pollen in *Rubus*. Bot. Gaz. 62: 370-388.
2. Brainard, E., and Peitersen, A. K., 1920. Blackberries in New England. Their classification. Vt. Agr. Exp. Sta. Bul. 217.
3. Peiterson, A. K., 1921. Blackberries in New England. Genetic status of the plants. Vt. Agr. Exp. Sta. Bul. 218.
4. Longley, A. E., 1924. Cytological studies in the genus *Rubus*. Amer. Jour. Bot. 11: 249-282.
5. Longley, A. E., and Darrow, G. M., 1924. Cytological studies of diploid and polyploid forms of raspberries. Jour. Agric. Research 26: 737-748.
6. Longley, A. E., 1926. Chromosomes and their significance in strawberry classification. Jour. Agric. Research 32: 559-568.





# SEED PRODUCTION IN STERILE CITRUS HYBRIDS —ITS SCIENTIFIC EXPLANATION AND PRACTICAL SIGNIFICANCE

WALTER T. SWINGLE

*Bureau of Plant Industry, U. S. Department of Agriculture*

Sterile first-generation hybrids are not uncommon especially when very diverse species are crossed. It is, however, unusual to find such hybrids producing seeds apparently normal in appearance and of good viability. Such seeds are produced by many sterile  $F_1$  hybrids obtained by crossing the common sweet orange, *Citrus sinensis*, with the Chinese trifoliate orange, *Poncirus trifoliata*.

The author made the first accurately controlled cross-pollinations between these two species and secured one hybrid, named the Rusk citrange (1 and 2), by using pollen of *Poncirus trifoliata* on a flower (bagged and castrated) of *Citrus sinensis*. Only one hybrid was produced by this crossed fruit. At the same time another cross was made by applying pollen of the sweet orange to a properly bagged flower of the trifoliate orange and from a crossed fruit so secured a dozen hybrid seedlings developed, among them the Colman, Morton, Savage, Rustic, Etonia, Norton, Phelps and Sanford citranges. These citranges have been under observation for more than a quarter of a century and second generation plants grown from the seed have been studied more than twenty years.

Most of the citranges have never yet produced a fertile female gamete and consequently have not yielded any second generation seedlings, although most if not all of them do produce some fertile pollen which can be and has been used in making complex hybrids with other citrus fruits such as the citrangequats which were obtained by using pollen of citranges on the flowers of the oval and round kumquats, *Fortunella margarita* and *F. japonica* (3).

In the course of very extensive cross-pollination work by the author and his colleagues it was soon found that all of the citranges set seeds if pollinated and that these seeds were usually plump and viable, but the seedlings were, in most of the citranges, exactly like the mother parent and showed no trace of any influence of the male parent. In the case of the Rusk citrange, which seeds rather freely, it was found that the seedlings (often grown many hundreds at once under identical conditions in the greenhouses) were remarkably similar in appearance. One such lot of Rusks, more than 1,500 in number, was carefully studied and failed to show any noticeable variation; the whole bench full looked exactly like plants propagated vegetatively. Now

that the Rusk citrange is being used on a considerable scale as a stock upon which to bud other citrous fruits, it is propagated both from seed and by cuttings and it is very difficult, if not impossible, to distinguish the cuttings from the seedlings when once the seedlings have gone past the early stage of germination. At the early stage, of course, they look very different from cuttings, which are set out with fully grown leaves. Many Rusk citrange seedlings have been fruited and in all such cases so far observed the fruit is exactly like that of the parent variety. As the Rusk is a very striking fruit, of deep red color when ripe, with an unmistakable aroma and flavor, it would be easy to detect any variation from the  $F_1$  in the seedlings. No such variations have been found, in spite of repeated search.

The Rusk citrange, and in fact most of the citranges, shows extraordinary vegetative vigor. Almost all of the citranges grow faster than the combined growth of both parent species. Several of them, and in particular the Rusk, have been found after long experience and tests to be excellent stocks upon which to graft the Satsuma and other oranges. It happens, therefore, in this case that the unusual and unexpected possibility of propagating generation after generation from seed without any perceptible variation is a very decided advantage in the practical utilization of these plants for stocks. Several of the citranges, and in particular the Savage, Morton and Rustic, all of which, like the Rusk, produce only  $F_1$  seedlings, are also being grown in the same way from seed for use as stock plants. Only two citranges, the Phelps and the Sanford, are known to produce regularly numerous true  $F_2$  seedlings.

The scientific explanation of this very curious and unexpected phenomenon is very simple indeed, as Strasburger (4) showed years ago the supernumerary embryos found in the seeds of most species of *Citrus* are produced by buds growing out from the nucellar tissue into an embryo sac. Naturally these buds reproduce the mother variety almost without variation, in the same way as do buds taken from ordinary vegetative branches. It is true that such nucellar buds do differ from ordinary buds in that the ontogeny is greatly changed and the young seedling developed from the nucellar bud has cotyledons and develops exactly like a normal seedling. This can perhaps be considered as a result of the powerful direct influence exerted on ontogeny by the embryo sac and its various organs. The changed ontogeny of such a nucellar bud can be compared somewhat remotely to the changed development of the egg or young larvae of the honey bee when the workers find that they have lost their queen and by enlarging the cell about the young larvae that would have developed into a worker cause it to develop into a queen bee by giving it special food and larger space for development. These and other relations observed in the development of nucellar bud embryos are valuable material for the study of endocrinology, until recently the monopoly of animal physiologists, but also beyond doubt of great importance in plant physiology and plant ontogeny as well.

It is known that supernumerary embryos derived from the mother plant

occur in mangos and in other plants; it is probable that they may likewise prove interesting both from scientific standpoint and in practical plant culture.

#### LITERATURE CITED

1. Webber, H. J., and Swingle, Walter T. New *Citrus* creations of the Department of Agriculture. U. S. Dept. Agric. Yearbook. 1904.
2. Swingle, Walter T. Variation in first generation hybrids (imperfect dominance): its possible explanation through zygotaxis. IV<sup>e</sup> Conference Internat. Génétique, pp. 381-394. Paris, 1913.
3. Swingle, Walter T., and Robinson, T. Ralph. Two important new types of *Citrus* hybrids for the home garden—Citrangequats and Limequats. Jour. Agric. Research 23: 229-238. 1923.
4. Strasburger, E. Ueber Polyembryonie. Zeitschr. f. Naturwissen-sch. 12: 654-678. 1878.





# THE COURSE OF POLLEN FORMATION IN CERTAIN ROSES, WITH SOME DEDUCTIONS THEREFROM\*

J. W. HESLOP HARRISON and K. B. BLACKBURN

*Armstrong College, University of Durham, Newcastle-on-Tyne, England*

During the past few years much of our research work has been planned with a view to elucidating the problems presented by so-called critical plant genera. Of these, to none have we devoted more time and study than to the genus *Rosa*. This genus, by the enormous spread of variation within its different groups, has practically defeated all attempts to classify it into species of the same value as those of other genera.

Indeed, so hopeless in some respects is the position that, in spite of the efforts of rhodologists of various countries, we have gone little beyond the position taken up by Linnaeus when he said: "Species rosarum difficillime distinguuntur, difficilium determinantur; mihi videtur naturam miscuisse plures vel lusu ex uno plures formasse; hinc qui paucas videt species facilius eas distinguit, quam qui plures examinavit."

To determine the causes of this uncertainty we have attacked the problem from various angles: that of the experimental breeder, of the field worker, of the parasitologist, of the cytologist, and so on.

In the present paper we propose more especially to deal with the results of our cytological investigations, although we shall not hesitate to utilize any useful facts yielded by our other work.

In our cytological examination of the *Rosae* we soon found that there existed in the roses a polyploid series based on the chromosome number of seven. More remarkable than this was the discovery that, in addition to forms carrying as their somatic chromosome complement an even multiple of the fundamental number, there existed others just as certainly endowed with odd multiples of that figure; of such forms we have recognized triploids, and pentaploids.

The behavior of roses of this latter type during meiosis could not be other than anomalous, and in the anomalies we see the explanation of much of the variability of the *Caninae* group of the Palaearctic roses. On the other hand, we naturally expected to find that the diploid, tetraploid, and hexaploid forms, or, as we prefer to call them, microgenes, would pass through all the stages of microspore formation on ordinary lines. This held true of the diploids, but, to our astonishment, we found that the tetraploids and hexaploids were of two types, one with pollen development of the usual type and the other in which it harmonized with the state of affairs in the triploids and pentaploids.

---

\*Presented to the Conference by Dr. Kathleen B. Blackburn.

For instance, although *Rosa pimpinellifolia*, *R. humilis*, and *R. lucida*, as well as the whole of the *Rosa mollis-omissa* group were tetraploid, in their meiotic behavior the former set agreed with the quite ordinary diploid *R. arvensis* and *R. rugosa*, whilst the latter, except in chromosome number, could not be distinguished from the anomalous pentaploids of the sections Eucaninae, Tomentosae and Rubiginosae. Similarly, with the hexaploids, although *R. acicularis* was quite ordinary, the meiotic process in *R. Sabini* followed substantially the lines of the pentaploids.

To distinguish these two types we applied the term "balanced" to those displaying normal behavior in the reduction division in the pollen mother cells; on the contrary, those in which the course of events was irregular we regarded as "unbalanced." Thus, we spoke of "balanced" and "unbalanced" tetraploids, of "balanced" and "unbalanced" hexaploids.

Next, we shall be asked in what respect is pollen development abnormal?

To discuss this adequately we must digress a little and glance for a while at the classical case of Rosenberg's *Drosera* hybrids. In that work Rosenberg dealt with the two species *Drosera longifolia* and *D. rotundifolia* and with their hybrid *D. obovata*.

He found that the two parent forms differed in chromosome number, *Drosera rotundifolia* having 10 chromosomes as its reduced number and *D. longifolia* having 20; the hybrid showed 30 as its somatic number. Regarding 10 as the base, this makes the hybrid a triploid form, and it becomes significant from the standpoint of the orthoploid series in *Rosa*.

During meiosis little cause for remark existed in the pure species, but in the hybrid only 10 bivalents appeared on the heterotype spindle with 10 univalents more or less irregularly disposed. At the anaphase the bivalents separated normally, whilst the univalents wandered, split, disappeared in the cytoplasm and so on. Thus, when the pollen tetrads should have appeared in the usual way, owing to these irregularities, many of the grains carried an abnormal chromosome complement and therefore collapsed.

In the triploid, pentaploid and unbalanced tetraploid roses much the same phenomena are to be observed. On the heterotype plates we find seven pairs of bivalents with the other chromosomes scattered irregularly about, either on the plate or elsewhere on the spindle. These bivalents proceed to the anaphase long before the univalents, which, as in the *Drosera* hybrid, wander, lag on the spindle and show all possible degrees of abnormal behavior. Thus, in many cases, only seven chromosomes appear on the homotype plates, to result subsequently in microspores carrying a nucleus built up of 7 chromosomes, and usually only these pollen grains are fertile. Indeed, often enough, when many chromosomes fail to reach the pole in the heterotype division, supernumerary micronuclei are generated which, also dividing at the homotype stage, result in the appearance, not of pollen tetrads, but of "octads" and similar groups in which most of the grains fail to mature.

Comparing the occurrences in the Canine roses with the chromosome behavior of the recognized hybrid *Drosera obovata*, and coupling this with the

fact that *R. Sabini* (the cytology of which resembles that of the pentaploids) is a patent hybrid between *R. pimpinellifolia* and some members of the Tomentosa group, we cannot escape from the conclusion that the Canine roses are themselves of hybrid origin.

Recognizing this, and also that the group is dominant in Europe, we next ask how it maintains itself.

In the first place let us look at the pollen conditions in a series of Canine microgenes. These yield the following percentages of good pollen:

#### 0 TO 10 PER CENT

AFZELIANAE: *R. subcristata*, *Reuteri*, *subcanina*, *fugax*.

EUCANINAE: *R. inconspicua*, *biserrata*.

RUBIGINOSAE: *R. echinocarpa*.

AGRESTES: *R. Borreri*.

VILLOSAE: *R. coerulea*, *pseudorubiginosa*.

TOMENTOSAE: *R. tomentosa*.

#### 10 TO 30 PER CENT

AFZELIANAE: *R. frutetorum*, *coriifolia*, *venosa*.

EUCANINAE: *R. lutetiana*, *hemitricha*, *aciculata*.

RUBIGINOSAE: *R. comosa*.

TOMENTOSAE: *R. sylvestris*, *pseudocuspidata*, *foetida*.

#### 30 TO 50 PER CENT

EUCANINAE: *R. flexibilis*.

RUBRIFOLIAE: *R. rubrifolia*.

VILLOSAE: *R. submollis*.

#### 50 TO 70 PER CENT

EUCANINAE: *R. fallens*.

VILLOSAE: *R. omissa*.

#### 75 TO 90 PER CENT

EUCANINAE: *R. senticosa*.

VILLOSAE: *R. mollis*.

Admittedly, whilst most of these show a high degree of pollen sterility, some of these microgenes produce enough sound pollen to be more or less effective; others, however, do not. How then is reproduction effected? That some method of securing it does exist is speedily proved.

Take, for example, *R. subcristata*, *R. fugax* and *R. coerulea*, in all of which the whole of the pollen aborts. Visit the shrubs in September, and no rose makes a braver display of crimson globes than they. It looks clearly as if they relied on some process other than that of normal fertilization. This we tested experimentally by castrating many plants of the Eucaninae, Villosae and Rubiginosae and bagging them. Without exception, seeds were set; hence the Canine roses, to say the least, are facultatively apomictical, and this has a definite bearing on the question of their origin.

In 1918 Ernst published a lengthy treatise, theoretical in the main, in

which he advanced the view that hybridity and apogamy are linked in the way of cause and effect. Though the evidence adduced points in that direction, in the absence of indisputable proof this is little more than a pious expression of opinion.

Recently, however, we have been able, as a result of hybridity experiments with lepidoptera, to demonstrate that parthenogenesis does arise in the  $F_1$  hybrids between *Tephrosia crepuscularia* and *T. bistortata* (Harrison and Peacock, 1925). The occurrence of apimixis is thus another link in the chain of evidence connecting the conditions of hybrids with those of the Canine roses.

Assigning due weight to the meiotic and other facts in the triploid, pentaploid, and unbalanced tetraploid and hexaploid roses, we are inclined to think that they have arisen as hybrids. There would thus be a perfect agreement with the usually accepted theories as to the development of polyploid series in plant genera. But how do we account for the higher balanced members of the series?

Clearly, if seven be the base chromosome number in *Rosa*, and diploid species occur, some of them must be more or less primitive and must have participated in the events which gave rise to the polyploids; hence we must look about for some process in which they can play a part. The simple crossing of two diploids would yield a diploid  $F_1$ , which, with a pairing of homologous chromosomes in the meiotic phase, brings us no further forward. However, we have direct evidence to offer as to the exact methods by which polyploids arise in *Rosa*. *Rosa Wilsoni* is a hybrid between *R. pimpinellifolia* and *R. tomentosa*, with the latter acting as pollen parent. Theoretically, therefore, it ought to have  $14 + 7$  chromosomes as its somatic number. Instead, direct cytological investigation shows it to be endowed with 42. Obviously, chromosome doubling by some means or other has occurred, but, what is most noteworthy, although the reciprocal hybrid is unbalanced and sterile, this is balanced and fertile. Thus we have generated before us, by the union of an egg with 14 chromosomes and a pollengrain with 7, a fully fertile hexaploid rose.

We, therefore, see not the slightest reason for rejecting in *Rosa* the usual views as to the hybrid origin of orthoploid series in plant genera.

If the fertile hexaploid arises thus, then the fertile tetraploid can be developed by a similar doubling in a cross between two diploids. In that case, once again, the supplying of homologous chromosomes would end in the attainment of fertility—and so with other members of the series.

Further, we believe that such happenings have taken place more than once in *Rosa*, for in the various sections the polyploid chains are quite independent. Take for example, the Cinnamomeae; starting with the diploid *R. cinnamomea*, we have the tetraploid *R. pendulina* and the hexaploid *R. nutkana*; in the Carolinae the diploid *R. nitida* and the tetraploid *R. lucida* and in the Pimpinellifoliae the diploid *R. Hugonis* and the tetraploid *R. pimpinellifolia*.



Recently, Hurst, arguing from the same series of observations as ours, has arrived at vastly different conclusions as to the development of such series. Instead of believing in the synthesis of the polyploids from lower members of the series, he regards all as having been derived from a theoretical decaploid polar species by the successive losses of chromosome septets. This view seems to us untenable and the reasons for our opinion are as follows:

Leaving on one side the parallelism existing between the conditions in *Rubus*, *Crataegus* and *Rosa*, we fail utterly to see why the same explanations offered and accepted as satisfactory for other plant groups like *Chrysanthemum* and *Campanula* should be rejected here.

One always regards, in matters numerical, the simple as the more primitive and the complex as the more advanced, yet here we are asked to believe that the decaploid existed first. This, as we have indicated, is in itself very improbable, but in addition is far from agreeing with chromosome numbers not only in primitive Rosaceae but also in the Leguminosae—an order, like the Rosaceae, a member of the cohort Rosales. There, likewise, a frequent chromosome number is 7 as in the genus *Lathyrus*, and we should rather imagine the two orders as making contact in their more primitive and fundamental numbers.

Again what is the most primitive species of *Rosa*? And what is its chromosome complement? The answer is *Rosa persica*—a diploid—primitive in respect to its simple leaves and the structure of its fruit. And with which other group does this primitive form link up? Unmistakably, it is closely allied morphologically to the Cinnamomeae, which are thus determined as forming a very early group indeed.

As we have seen already the Cinnamomeae possess in themselves a reasonably complete polyploid series and, furthermore, are a very homogeneous group of species well represented in Europe, Asia and America. In America we find only Cinnamomeae forms (with the doubtfully distinct Carolinae) and in the Palaearctic area, in addition to Cinnamomeae, our plethora of forms and groups.

It seems very unlikely that this distribution should have arisen by "defects" from a circumpolar decaploid species, but closer study of the circumstances attending the distribution of the Cinnamomeae still further lessens the plausibility of the view. Apart from the fossil evidence, which shows *Rosa* to have come into being in early Tertiary times, the geographical distribution of the Cinnamomeae is very significant. Comparing it with that of the less advanced members of the lepidopterous subfamily Bistoninae, which has been submitted to intensive study (Harrison, 1916), we find it to coincide with that of the twin genera *Amphidasys* and *Biston*, or, to take plant genera, with that of the Farinosae group of the genus *Primula*. We have, for very many reasons, assigned to *Amphidasys* and *Biston* a northeastern Asiatic origin, and most observers assume for *Primula* the same metropolis. Hence we imagine the Cinnamomeae to have originated in the same area and

to have spread thence, giving rise in America to *R. blanda*, *R. pratincola*, *R. arkansana*, etc., and in the Palaearctic region to *R. Sweginzowii*, *R. pendulina*, *R. setipoda*, *R. rugosa*, *R. cinnamomea*, etc. Many rhodologists regard the other sections as having been evolved from this group by mutation, by response to environmental influences or by hybridity. We were probably the first to state the latter view in the case of the Eucaninae and Rubiginosae, but Boulenger has extended it to include the Rubrifoliae, Villosae and Tomen-tosae.

We are inclined to think that the American Cinnamomeae have had a dual origin, regarding *R. nutkana*, not as a form derived by chromosome loss from *R. acicularis* as it moved south, but as a very recent immigrant to Pacific America and therefore comparable with the butterflies *Papilio machaon* and *Thecla (Callophrys) dumetorum*. Hence its origin (and possibly that of other western Cinnamomeae) is quite independent of the eastern representatives of the Section. Of this we have rather neat independent proof.

Of the gall-making groups attacking *Rosa* none is more characteristic than the Cynipid genus *Rhodites*; wherever roses exist there we meet with that genus. But the American forms, except for one species, form a very homogeneous group. And what is this exception?—*Rhodites bassetti* closely allied to our common Palaearctic *Rhodites rosae* and forming a very similar gall, confined to *R. nutkana*!

Hurst states in discussing this matter of geographical distribution, "In fact a general statement might be made that towards the Pole the number of septets of chromosomes and characters increases, while toward the equator the number decreases (Fig. 175)." Turning to this map, we see pictured a state of affairs, which, if accurate, would warrant the statement.

It does supply the distribution of *Rosa* as a whole and thereby confirms our opinion as to the Eastern Asiatic origin of *Rosa*. In addition it suggests an early migration of the Cinnamomean roses with later wanderings of other groups which took the usual routes of members of the Siberian and Oriental migrations passing into Europe and N. Africa.

However, on testing the accuracy of the various details of the map we find that they do not agree with the facts, the forms high in polyploid series all being placed much too far north and, on the contrary, the diploid species uniformly appearing too far south. A semblance of harmony is thus produced between the distribution and the main thesis.

To take actual details of the map, we find octoploid forms figured as occurring far to the north in America, Iceland, Europe and Siberia. This is erroneous in several respects. In the first place, it assumes that *Rosa acicularis*, the rose inhabiting the Holarctic region, is octoploid throughout. Admittedly Täckholm found an octoploid *R. acicularis* in Europe, but this does not alter the fact that he also examined hexaploid forms of the same species. Apart from this, Penland and ourselves, the former from United States material and ourselves with material from the northern Manitoba-Saskatchewan border, found the American plant to be hexaploid. Moreover, the

American distribution indicated is much too restricted, for *R. acicularis* has been collected by ourselves in Saskatchewan, Manitoba and Ontario, whilst its occurrence is reported from Michigan (Erlanson) N. Y. (Standley) and Texas (Schneider). Similarly, the figure "8" is placed in Iceland to indicate that *R. acicularis* grows there, but, as is well known, the only rose there is the tetraploid *R. pimpinellifolia*!

Turning next to the Mediterranean area, we note in Africa only the figure "2," the same figure labelling the rose flora of S. Spain, Italy and Sicily. Once more the map is not in accordance with the facts, the roses in the Canaries, Madeira, N. Africa, Italy, Sicily, S. Spain being predominantly pentaploid.

Submitting now the Asiatic distribution to critical examination, and testing it on the diploid *Rosa rugosa*, we find it represented as stopping short in Korea and S. Japan—although this rose is well known from Kamchatka more than a thousand miles to the north.

Although the list of mistakes like these could be lengthened almost indefinitely, we shall confine ourselves to one more example—a case from our own islands. Here we see situated in the north of England and Ulster the figure "6" which implies that we possess a hexaploid microgene. This we deny completely; we do certainly possess hexaploid forms, but these are obvious hybrids between tetraploids and unbalanced pentaploids. How casual hybrids, liable to be produced at any time, can have any bearing on the declared purpose of the map we utterly fail to see.

Let us now consider what support the cytology of the microgenes affords this theory of Hurst's.

To simplify the discussion, let us adopt that worker's symbolical representation of his theoretical circumpolar decaploid. To it he assigns the formula AABBCDDDEE, each capital denoting one of his postulated differential chromosome septets; in the same way his supposed derived diploids have the formulae *AA*, *BB*, *CC*, *DD*, and *EE*, respectively.

On this basis he represents *Rosa* "*canina*" as *AABDE*, thereby indicating (1) that it possesses only one set of the septets *B*, *D* and *E*, but (2) that the *A* set is duplicated. This, of course, is intended to be in agreement with the fact that on the heterotype spindle during pollen formation in this group of species there are seven bivalent and 21 univalent chromosomes. It, however, implies that the individual septets will only pair up with others of the same order as themselves; *A* with *A*, *B* with *B* and so on.

This cannot be correct, as is easily shown. For instance, in the *pimpinellifolia* × *canina* hybrids, where a species *BBCC* (on this notation) is crossed with a form *AABDE*, in pollen formation 14 chromosomes find mates. This, accepting as correct Hurst's position, would mean that, in addition to the two *BB* sets of the hybrid pairing as one would anticipate, the *C* introduced by *R. pimpinellifolia* had paired up with septet *A*, *D* or *E*, a very improbable thing if the "septets" exist, and are differential. Much the same kind of evidence is provided by the *rugosa-cinnamomea*, *nutkana-pendulina*,



*lucida-rugosa*, *cinnamomea-pendulina*, and the multitudinous *chinensis* crosses.

Such occurrences seem much more in accord with a hybrid origin for the orthoploid series, and allow for varying shifts of values as the various diploid, and tetraploid species diverged physiologically from the species from which they were evolved.

Let us consider now Hurst's own cytological discussion as to the origin of *Rosa* species.

He begins by formulating the cytological difficulty that the union of two of his differential diploid species *AA* and *BB* could only result in a sterile diploid *AB*. Alternatively, he assumes that, if fertile, such a hybrid could only give rise to plants of its own build and plants like its parents of composition *AA* and *BB*. We can only say that, not only is the sterility of such hybrids "not proven," but, on the contrary, in our experience, and in that of rose growers generally, the majority of such hybrids are fertile. Further, we ask for a single trace of proof that the chromosomes in diploids, instead of acting independently as in other plants, move in blocks of seven. We have looked seriously for evidence of such behavior but have never been able to detect it.

Moreover, the weightiest evidence we can adduce, i.e., the case of *Rosa Wilsoni*, demonstrates conclusively that the unlikely (in Hurst's opinion) event of chromosome duplication does occur, and does result in fertility. Hence we consider that the production of an *AABB* form from *AA* and *BB* parents a far from unlikely contingency. In addition, in our opinion gratuitously, he asserts that such a duplication gives rise to a homozygous tetraploid species whilst all the tetraploid species of *Rosa* are certainly heterozygous. Granting that the *Rubrifoliae* and *Villosae* are heterozygous tetraploids, what about *R. pimpinellifolia*, *R. pendulina*, *R. humilis*, *R. lucida* and the other balanced tetraploids, all necessarily homozygous? Even Hurst himself concedes the homozygous nature of *R. spinosissima* and *R. altaica* by writing their formulae as *BBCC* and *BBDD*, respectively.

To buttress up his case against the synthetic nature of rose polyploids he raises the geographical difficulty that the species necessary to generate polyploids are not present in the same stations to hybridize. Allowing for the immense age of the genus, we should indeed be surprised, with all the movement of plant species that has taken place, if such were so. This argument is much on the same level as supposing that Hurst's own theoretical decaploid, and all its possible "defect" species, should yet exist where they originated!

Next he propounds the view that the evolution of the Arctic polyploids, by the hybridization of subtropical diploids, implies a distinct reversal of accepted opinions as to the Arctic origin of the Flora of the Northern Hemisphere. Once again we have two assumptions stated as facts: (1) that *Rosa* is Arctic in origin, and (2) that the diploids are subtropical; in other words Hurst is here arguing in a circle. We have already put forward our ideas as to the origin of *Rosa* whilst the second point concerning the "subtropical"



nature of the diploids rests simply on his map and is not warranted by facts. We ourselves have collected diploid *R. blanda* far north in Canada; diploid *R. rugosa* goes up to Kamchatka in Asia, diploid *R. arvensis* reaches Scotland, diploid *R. cinnamomea* is more characteristic of Scandinavia than of Southern Europe and so on.

Again "an origin from five primitive diploids," he asserts, "implies a multiple origin with its consequent ulterior problems." We cannot agree with this. In species after species of *Silene* the reduced chromosome number is 12; in many Lepidoptera it is 31 and in Orthoptera 12. No inherent difficulty lies there to a multiplicity of diploid species; why then should we invent one for *Rosa*? The derivation of the diploids from a primitive polyploid is therefore not a more simple proposition.

We see a further objection to Hurst's hypothesis in the assumption that each individual septet of chromosomes in the original decaploid bears all the potentialities of the rose species. This seems an extremely improbable situation, and entirely at variance with all the pertinent facts, for, whilst we do know that each multiple of the base number does carry the whole powers in the case of a tetraploid *Primula*, we are likewise certain that such a *Primula* arose by a duplication of the pre-existing complex.

To sum up: as a direct deduction from the pollen sterility and the course of its development in the unbalanced roses, we conclude that such roses originated in hybridity and, secondly, from allied considerations in the case of *R. Wilsoni*, we regard the balanced polyploids as of similar origin attaining fertility simultaneously with, and as a direct consequence of, a doubling of their chromosome complements. Thus we would seek to account for the orthoploid series developed in the various sections into which the roses are divided.

[I have to acknowledge a grant from the Armstrong College Research Endowment Fund Committee in aid of the work discussed above. J.W.H.H.]

#### BIBLIOGRAPHY

- Blackburn, K. B., and Harrison, J. W. Heslop, 1921. The status of the British rose forms as determined by their cytological behavior. *Ann. Bot.* **35**: 159-188.
- Blackburn, K. B., and Harrison, J. W. Heslop, 1924. Genetical and cytological studies in hybrid roses. 1. The origin of a fertile hexaploid form in the Pimpinellifoliae-Villosae crosses. *Brit. Jour. Expt. Biol.* **1**: 557-570.
- Boulenger, G. A., 1924-5. Roses d'Europe. Brussels.
- Erlanson, E. W., 1925. The wild roses of the Mackinac Region of Michigan. *Papers Mich. Acad. Arts and Letters* **5**: 77-94.
- Ernst, E. W., 1918. Bastardierung als Ursache der Apogamie im Pflanzenreich.
- Harrison, J. W. Heslop, 1916 and 1917. The geographical distribution of the moths of the subfamily Bistoninae. *The Naturalist* **1916**: 163-166, 194-198, 273-278, 358-362, 377-382; and **1917**: 161-164, 252-257, 293-296, 312-320.
- 1921. The genus *Rosa*, its hybridology and other genetical problems. *Trans. Nat. Hist. Soc. Northumberland, etc.*, **II**: **5**: 244-298.
- Harrison, J. W. Heslop, and Peacock, A. D., 1926. On parthenogenesis originating in lepidopterous crosses. *Transactions Natural History Society of Northumberland, Durham and Newcastle-on-Tyne. (New Series.)* **6**: Part II, 201-217.
- Hurst, C. C., 1925. Chromosomes and characters in *Rosa* and their significance in the origin of species. In "*Experiments in Genetics*," 534-550, Cambridge.

- Kinsey, A. C., 1922. Studies of some new and described Cynipidae (Hymenoptera). Indiana University Studies 9: 3-141.
- Linnaeus, 1753. Species Plantarum.
- Longley, A. E., 1924. Cytological studies in the genus *Rubus*, and Cytological studies in the genus *Crataegus*. Am. Jour. Bot. 11: 249-282; 295-317.
- Marchal, E., 1920. Recherches sur les variations numériques des chromosomes dans la série végétale. Mem. Acad. Roy. de Belgique, Series 2. 4: 1-198.
- Penland, C. W. T., 1923. Cytological behavior in *Rosa*. Bot. Gaz. 76: 403-410.
- Rosenberg, O., 1909. Cytologische und morphologische studien an *Drosera longifolia*  $\times$  *rotundifolia*. K. Svenska Vetensk. Akad. Handl. 43: 1-63.
- Schneider, C. K., 1906. Handbuch der Laubholzkunde. Vol. 1.
- Standley, P. C., 1921. Flora of Glacier National Park, Montana.
- Täckholm, G., 1922. Zytologische Studien über die Gattung *Rosa*. Acta Horti Bergiani 7: 97-381.
- Tahara, M., 1921. Cytologische Studien an einigen Kompositen. Journ. Coll. Sci. Tokyo 43: 1-53.

## DORMANCY IN HYBRID SEEDS

WILLIAM CROCKER

*Boyce Thompson Institute for Plant Research*

In speaking of the germination of hybrid seeds today, I want to emphasize mainly the germination of hybrid seeds of Rosaceae of the colder north temperate zone. This family of plants is of especial interest to the members of this Conference, for it includes so many of our fruit and ornamental plants. Of the many inquiries we receive concerning the germination of seeds, those regarding the rose family are most common.

The seeds of three sub-families (Pomeae, Roseae and Pruneae) have been studied in some detail. The seeds of every genus studied to date need a period in germinative conditions at a low temperature to prepare them for germination, or to after-ripen them. The old practice of stratifying certain seeds at a low temperature in moist condition preparatory to planting, furnished this condition in a general way. This practice often failed, in part, because it was not realized that the process of after-ripening in these seeds had a rather definite temperature optimum. Our work has established beyond a doubt the existence of such temperature optima and that the optima may vary somewhat with different species, although it is more generally in the region of 5° C. In some cases the optima are very distinct or involve a narrow range of temperature. In other cases the optima are not so distinct or represent a rather wide range of temperatures. Our work has also established that the various species have a distinct after-ripening time at their respective temperature optima, that a good oxygen and water supply is important in the stratification beds and that it is important to regulate the acidity of the stratification medium in some cases. In our experiments we mix the seeds up with the stratification medium instead of using successive layers of seeds and stratification medium.

The accompanying figure on seeds of *Rosa rubiginosa* shows the existence of the optimum. The after-ripening occurs at 0° C. and less readily at 10° C., but the process requires a much longer time at either temperature than at 5° C. At temperatures much above 10° C. the seeds tend to go back into the dormant condition so a period at higher temperatures prolongs the necessary time for after-ripening. Periods at temperatures below freezing have no effect on after-ripening provided they are not low enough to give freezing injuries. Contrary to the general view, freezing any of the rosaceous seeds of the three sub-families mentioned does not aid after-ripening in any way so far as our studies indicate. It does not even assist in cracking the stone coats (7). The stone coats of the plum, for instance, crack quickest at 10° C. or a little above, although the after-ripening occurs quickest at lower temperatures. There is no doubt that some seeds are much aided in germination by

freezing or repeated freezing, but so far as now certainly known, these are not seeds that demand an after-ripening of the embryos.

*Rosa multiflora* requires only a little over two months at 5° C. for after-ripening while *Rosa canina* shows considerable germination under this condition only after nearly a year of favorable stratification at this temperature. The study of these and many other species of roses of the cold temperate zone indicate that the various species demand various periods of cold stratification for after-ripening but in general the optimum is near 5° C.



FIG. 1. *Rosa rubiginosa* seeds: Check stored dry; the others in moist sand for six months at the temperatures designated and then planted in a flat in the greenhouse. The picture shows the effective temperature for stratification, that is 5° C.

We have also studied the after-ripening of about two hundred and fifty different crosses of roses. The indications from these studies are that hybrids containing largely cold temperate blood respond to low temperature stratification as do the seeds of the species with about 5° C. the optimum, and with a great variation in the time required. Apparently with hybrids of warm climate forms, the low temperature stratification is not so important, if at all necessary. It is probable that the low temperature stratification is merely an imitation of Nature's method for temperate zone forms. One can improve on Nature, however, for his purpose by giving the most favorable conditions as to temperature, time and stratification medium. Nature's purpose is achieved if a few of each year's crop of seeds germinate at a time over a period of years, even with a low total yield. The hybridizer wants a big yield as promptly as possible, hence he must improve on Nature's method.

TABLE 1 shows the effect of 0°, 5°, and 10° C. stratification upon the germination of Damson plum seeds both in and out of stone coats. These seeds do not germinate readily at 5° C. or below, but if fully after-ripened at



5° C. they do germinate readily when transferred to higher temperatures. It is evident from the table that 5° C. is a much more favorable stratification temperature than either 0° or 10° C. In these seeds the after-ripening occurs almost as rapidly when the stone coats are left on as when they are removed. One avoids much difficulty with molds by leaving the stone coats intact. These seeds after-ripen quite as readily in granulated peat ordinarily used by nurserymen, whether in its natural condition with a pH of 4.5 to 5 or leached with water giving a pH of about 6. It is much easier to control the moisture in peat than in sand.

TABLE 1

DAMSON PLUM SEEDS KEPT AT LOW TEMPERATURES IN MOIST SAND FOR 12 AND 16 WEEKS AND THEN TRANSFERRED TO THE HIGHER TEMPERATURES INDICATED IN THE TABLE. STERILIZED WITH .25 PER CENT USPULUN FOR .5 HOURS

IN CARPEL	PER CENT GERMINATION AFTER	
	12 WEEKS STORAGE	16 WEEKS STORAGE
Seeds transferred from 0° to 15°.....	0	0
Seeds transferred from 0° to 20°.....	0	0
Seeds transferred from 5° to 15°.....	90	...
Seeds transferred from 5° to 20°.....	80	100
Seeds transferred from 5° to greenhouse.....	...	100
Seeds transferred from 10° to 15°.....	10	...
Seeds transferred from 10° to 20°.....	10	10
OUT OF CARPEL		
Seeds transferred from 0° to 15°.....	10	...
Seeds transferred from 0° to 20°.....	20	30
Seeds transferred from 5° to 15°.....	80	...
Seeds transferred from 5° to 20°.....	100	...
Seeds transferred from 5° to greenhouse.....	...	70
Seeds transferred from 10° to 15°.....	10	...
Seeds transferred from 10° to 20°.....	10	20

TABLE 2 shows the after-ripening and germination of peach seeds under various conditions. There is little difference in the effectiveness of 5° C. and 10° C., while 15° and 20° C. constant and alternations between -10° C. and + 10° C. all give no germination for the period of the experiment. Granulated peat and sand are almost equally good as stratification media which means that these seeds are indifferent to a considerable range of acidity. The removal of the stone coats hastens germination and increases the percentage of germination very markedly. The common idea that peach pits need freezing to aid germination is not justified by this experiment or many others that we have run. Hybridizers of peaches often get very low yields of seedlings from their hybrid seeds. They will do well to follow the methods indicated by this experiment.

TABLE 2

PEACH SEEDS COLLECTED FROM FRUITS IN LATE SUMMER, CLEANED THOROUGHLY AND STORED DRY UNTIL EXPERIMENT STARTED ON OCTOBER 6, 1925. STERILIZED IN .25 PER CENT USPULUN FOR .5 HOUR BEFORE STRATIFYING

TEMP.	MEDIUM	STONE COAT	PER CENT GERMINATION AFTER					
			5 WEEKS.	10 WKS.	14 WKS.	18 WKS.	22 WKS.	26 WKS.
5° C.	Sand...	Intact....	.....	..	12	16	..	20
	Sand...	Removed..	.....	56	92	96	..	..
	Peat...	Intact....	.....	..	12	..	20	28
	Peat...	Removed..	(4% moldy)	72	96	..	..	..
10° C.	Sand...	Intact....	.....	..	20	32	40	44
	Sand...	Removed..	(4% moldy)	40	88	..	..	..
	Peat...	Intact....	.....	..	4	8	8	12
	Peat...	Removed..	(16% moldy)	44	84	..	..	..
15° C.	Peat...	Intact....	.....	..	..	..	..	..
20° C.	Peat...	Intact....	.....	..	..	..	..	..
Weekly alternation between +10° and -10° C.....	Peat...	Intact....	Some seeds with split coats but no germination.					

TABLE 3 shows the effect of various conditions upon the after-ripening and germination of French pear seeds. These seeds do not show a very sharp temperature optimum for after-ripening and germination. They do almost equally well at 0°, 5° and 10° C. Other experiments indicate that peat is superior to sand largely because it gives a more regular supply of oxygen and water. A number of nurserymen have obtained very poor yields from French

TABLE 3

FRENCH PEAR SEEDS COLLECTED FALL 1925 AND STORED DRY UNTIL FEBRUARY OF 1926 WHEN THE EXPERIMENT WAS STARTED. STERILIZED IN .25 PER CENT USPULUN BEFORE STRATIFYING

TEMP.	MEDIUM	TREATMENT	PER CENT GERMINATION AFTER				
			11 WEEKS	13 WEEKS	15 WEEKS	17 WEEKS	19 WEEKS
0° C.	Washed peat...	Uspulun.....	...	.5	24.5	61.5	....
5° C.	Washed peat...	Uspulun.....	3.5	13.0	61.0	75.0	82.0
10° C.	Washed peat...	Uspulun.....	1.0	....	8.5	55.0	....
0° C.	Washed peat...	Not treated...	...	2.0	25.0	66.0	....
5° C.	Washed peat...	Not treated...	.5	8.5	18.5	57.0	70.0
10° C.	Washed peat...	Not treated...	4.5	....	7.5	37.5	....
5° C.	Sand.....	.....	transferred to peat	5.0	7.5	47.0	69.5

pear seeds that they grow for understock. Their troubles would probably be largely overcome if they stratified the seeds at low temperatures two or three months previous to early spring planting. Losses from molds are also greatly reduced by sterilizing the seeds before stratification with uspulun or semesan.

The results given in tables 1-3 are typical of the results obtained with many genera species and varieties of the three sub-families of Rosaceae mentioned above.

Various genera and species of several other families of plants respond to low temperature stratification as do the rosaceous seeds. Table 4 shows the effect of low temperature stratification upon the after-ripening of *Cornus nuttallii*. In this 0° C. is more rapid in its action than 5° C., and 5° C. is more rapid than 10° C., also peat is better than sand. Jones (3) has shown that fall fruiting maples respond to low temperature stratification and Rose (6) has shown the same for *Tilia americana* and Pack (4) the same for *Juniperus*. Our work has added several other genera of various families to this list.

TABLE 4

*Cornus nuttallii* FROM SEATTLE, WASH., HARVESTED IN THE FALL OF 1925. SEEDS WERE CLEANED AND STERILIZED WITH .25 PER CENT USPULUN FOR .5 HOUR. SEEDS SHOWED NO MOLD. EXPERIMENT STARTED SEPTEMBER 23, 1925

TEMP.	CONDI- TION	PER CENT GERMINATION AFTER							
		18 WEEKS	22 WEEKS	26 WEEKS	30 WEEKS	34 WEEKS	38 WEEKS	42 WEEKS	45 WEEKS
0° C.	Peat...	8.2	35.1	49.0	75.7	79.3	85.0	86.0	....
	Sand...	1.0	4.0	20.0	39.3	51.0	73.0	75.0	75.5
5° C.	Peat...	0.3	1.3	46.8	68.0	76.5	93.0	93.0	93.0
	Sand...	....	....	....	3.4	3.4	5.5	26.0	28.0
10° C.	Peat...	0.3	1.0	1.0	20.6	39.0	71.0	75.0	76.0
	Sand...	....	....	....	4.0	6.0	6.0	5.5	7.5

The chemical and enzymic changes occurring in seeds with dormant embryos in low temperature stratification are described by Pack (5) for *Juniperus* and Eckerson (2) for *Crataegus*.

In closing it should be emphasized that hybridizers of rosaceous forms will profit by making use of low temperature stratification in production of their hybrid seedlings.

#### LITERATURE CITED

1. Davis, Opal Hart. Germination of seeds of certain horticultural plants. Florists Exchange and Hort. Trade World. Nov. 13, 1926.
2. Eckerson, Sophia H. A physiological and chemical study of after-ripening. Bot. Gaz. 55: 286-299. 1913.
3. Jones, H. A. Physiological study of maple seeds. Bot. Gaz. 69: 127-152. 1920.
4. Pack, D. A. After-ripening and germination of *Juniperus* seeds. Bot. Gaz. 71: 32-60. 1921.

5. ——— Chemistry of after-ripening, germination, and seedling development of juniper seeds. Bot. Gaz. 72: 139-150. 1921.
6. Rose, R. C. After-ripening and germination of seeds of *Tilia*, *Sambucus* and *Rubus*. Bot. Gaz. 67: 281-308. 1919.
7. Tukey, H. B. Studies of fruit storage and germination. Bul. 509, N. Y. State Agr. Exp. Sta., Geneva. 1924.



# STERILITIES AND SEED PRODUCTION IN DAHLIAS

J. B. S. NORTON

*University of Maryland*

The ray flowers of the common double varieties of the dahlias of our gardens are generally without stamens and have various degrees of abortion of the pistil (see Gerard in Rivoire, *La Dahlia*). Some varieties, however, sometimes produce seed in the ray flowers.

Dahlias are grown from seed as annuals, especially the single types. Seed of the fine double varieties is much desired for producing new kinds. Practically all the seed is produced in the disk flowers. Many of the best double varieties are sterile or bear only a few seeds in the center of the head so that progeny of the most valuable kinds is difficult to obtain.

The short days of spring and fall stimulate dahlia flower-production and the development of more fertile disk flowers and fewer rays in the double varieties. Very double varieties can then be made to produce seed by growing early in greenhouses or by covering to reduce the day length.

Out of about 30 varieties grown and crossed and selfed in various ways in the last 3 years, in the greenhouse from February to May, only one certainly produced fertile seed with its own pollen. Patrick O'Mara with pollen from the same variety produced apparently good seed but they were without embryos.

Seed for producing new and improved varieties is generally taken from known mothers only, the pollen being supplied by bees. More careful workers have gone to much trouble and used many complicated methods of getting hand-pollinated and pedigreed seeds, and careful methods are necessary for genetical studies. But for all practical purposes, for the production of seed with both parents known, it is only necessary to plant the root clumps close together in the greenhouse in February or March, thus producing in a few weeks of short days, an abundance of disk flowers, rub the open flowers together daily, and keep the record of crosses on tags on each head. No emasculation is necessary, and unless greenhouse insects interfere, there is little danger of pollen from other sources, as no dahlias are in the gardens at that season, and their own pollen found in abundance over the blooming heads is rarely effective.

The seeds may be harvested when mature, or while still undried, planted, and flowers of the new cross secured in the field the same summer.

A frost-hardy dahlia would be a great acquisition to our gardens. *Bidens*, as indicated by leaf form and the occasional production of long, but not barbed, pappus horns on dahlia seeds, seems the most promising hardy form of dahlia relatives to use for putting cold resistance into dahlia crosses. So far, attempted crosses with three species of *Bidens* and two of *Coreopsis* have not been successful.

## THE ROLE OF STERILITIES IN THE BREEDING OF FLORICULTURAL PLANTS

A. C. BEAL  
*Cornell University*

A history of the development and improvement of plants would be very interesting if it could be written. Man probably early recognized differences in plants and tried to make use of the differences found in nature. At least this is the idea one gains from the writings of Theophrastus and Pliny. Theophrastus says that "Among roses there are many differences in the number of petals, in roughness, in beauty of color, and in sweetness of scent. Most have five petals but some have twelve or twenty, and some have a great many more than these; for there are some, they say, which are even called 'hundred-petalled.' Most of such roses grow near Philippi; for the people of that place get them on Mount Pangaeus, where they are abundant, and plant them. \* \* \* Some kinds are not fragrant or of large size. \* \* \* In general, as has been said, good color and scent depend upon locality. Sweetest scented of all are the roses of Cyrene."

Pliny writes that "the most esteemed kinds of rose among us are those of Praeneste and Campania." Some persons have added to these varieties the rose of Miletus, the flower of which is an extremely brilliant red and has never more than a dozen petals. Next to it is the rose of Trachyn, not so red as the last, and then that of Alabanda with whitish petals, but not so highly esteemed. Least esteemed of all, however, is the thorn rose, the petals of which are numerous, but extremely small. The essential points of difference in the rose are the number of the petals, the comparative number of thorns on the stem, the color and the smell. About twelve forms of roses were known to the Romans. It will be noted that the differences were usually associated with locality and hence probably represented species rather than varieties. Nowhere in the ancient world do I find the variety conception nor do we find much that indicates variation in ornamental plants under cultivation. It would seem that the ancients accepted the fortuitous variations flung in their path and since they were acquainted with all of the usual methods of propagation they could multiply them.

For about two thousand years—from Theophrastus to Clusius—plant improvement seems to have been practically stationary. The two ornamental plants which seem to have first given rise to many varieties were the tulip and the carnation. Clusius raised the first varieties of tulip during the closing years of the sixteenth century and the Dutch became so interested in the production of new varieties that the speculation known as the "tulip mania" arose during the seventeenth century.

Tulips and other bulbous plants were improved by Dutch gardeners but then, as now, the market for them was found in the neighboring countries. The oldest extant catalogues—those of the Morin brothers—indicate an interest in bulbous plants on the part of the French. Pierre Morin, 1651-1655, catalogued 100 varieties of tulips, 24 of ranunculus, 58 of iris, and 27 of anemone.

A century later another furor of similar, but of milder character developed over the production of hyacinths which were appearing in large numbers. A catalogue of the year 1767 enumerates 1670 varieties but it is doubtful if any are to be found in the extensive list of the present day.

While the Dutch were busy developing the tulip in hundreds of varietal forms, the French were interested in the oeillet or carnation. The first of the special books on one flower was the "Le Jardinage des Oeillet," by L. B., published 1647. About seventy varieties are mentioned. The second book on the carnation, unlike the first, is an eminently practical work and the anonymous writer describes the best varieties then known. There are about three hundred of these distributed by color as follows: 136 violet, 71 red, 33 incarnats, 2 rose, 6 white, 36 piquettes (picoties), 9 tricolor, a quadricolor and a quincolor. Truly a wide range of color. More significant were the ideals of a good carnation. The flowers should be large, furnished with numerous petals, round, well striped and without spots.

I have brought in this history because I want to show that it was in the seventeenth century that plant improvement really began, and I have mentioned the plants which received attention. Soon cultivators began to formulate ideals as to what constitutes beauty in their favorite flowers. The practice of holding flower shows, beginning in 1660 in England, naturally tended to diffuse these ideals. Mention should be made of the fact that many foreign plants reached Europe during the seventeenth century and the earliest greenhouse structure (1619), that the modern world knows anything about, was erected.

Early in the eighteenth century (prior to 1717) the first hybrid between a carnation and the sweet william was raised by Thomas Fairchild of Hoxton, England. Nevertheless it may be said that until recent times the practical gardener in his search for new varieties has relied on natural crossing. The great groups of *Rosa*, *Dahlia*, *Paeonia*, *Iris*, *Gladiolus*, probably arose in this way.

However, the work of Dean Herbert in the early part of the nineteenth century soon attracted attention. Possibly the first important hybrid, measured by its influence on modern gardening, was *Gladiolus gandavensis*, obtained in 1837 and considered by its raiser to *G. cardinalis*  $\times$  *G. psittacinus*. It seems certain, however, that it really is *G. cardinalis*  $\times$  *G. oppositiflorus*, the latter being grown by him and employed in some of his crosses.

The orchids because of their interest, rarity and value and because they must be carefully grown under glass under controlled conditions, have been much hybridized. The first artificial hybrid was flowered in 1856 and since



then great numbers have been raised. No other group can present as large numbers of hybrids of definite parentage. Here are to be found the largest number of generic hybrids.

About 1870 there seems to have come a change in the ideas of horticulturists regarding plant improvement. The work of Herbert together with the results of others doubtless attracted the attention of some enterprising spirits. Soon we had Henry Echford with sweet peas, Sir Michael Foster with irises and Henry Bennett with roses. In America we can point with pride to the development of the forcing type of carnation by Dorner, Starr, Fisher and others, and to the development of splendid climbing roses through the efforts of Manda, Walsh and Dr. Van Fleet.

We should recognize the fact, however, that nearly all of the important cut flower crops of the florist are perennial and that most of the annuals are relegated to the garden. The florist feels sure of the fact that, with most of the crops he grows, he can multiply by asexual methods and with more or less rapidity any desirable variation obtained by hybridization or mutation. So far as the perpetuation of varieties is concerned he is not much concerned whether they are unstable or sterile.

Enterprising florists and gardeners, however, are constantly seeking novelties for the improvement of a flower or to give it a wider range of usefulness.

It is at this point that the question of sterility assumes importance. Of course the producers of new varieties had long been aware of the fact that some varieties failed to set seed or that the seed was not viable, but records are not available. Probably the early improvement of tulips, hyacinths and other bulbs, referred to above, was from open fertilized seed. It is true of large numbers of Gallica, Hybrid China and Hybrid Perpetual roses sent out during the first half of the nineteenth century. We know the seed parent of many of the Hybrid Perpetual roses but not until the advent of the Hybrid Tea roses do we find many varieties of known parentage.

Naturally we expect through hybridization to raise the general level; otherwise the raising of seedlings will result in striking the same note over and over again. I think that one of the probable causes of the slump in the popularity of the carnation is due to the fact that while we can overcome the deterioration caused by forcing the plants, by the continued raising of seedlings, we nevertheless travel round and round a circumscribed circle. The carnation of today seems to be little, if any, improvement over that of twenty-five years ago. New varieties are in the nature of replacements and lack novelty.

When we come to analyze varieties for possible desirable characters for use as parents or when we attempt to use them we may be confronted with the conditions of sterility.

It should be pointed out in this connection that we should accept the older records of sterility with caution. The work of Knudson in germinating orchid seeds and that of Crocker with seeds in general would seem to indicate where

some of the early failures lie. Again we need to remember that while certain plants may be apparently sterile they may yield important results with some other form not hitherto tried with them. For many years the Persian Yellow rose did not yield results either as a pollen or seed parent. Finally Lord Penzance produced his sweet briers Lord and Lady Penzance, and still later this rose yielded the initial form of the increasingly important group of Pernetiana roses.

Among the important cut flowers in greenhouse and garden probably the most unusual is the *Lathyrus odoratus* or sweet pea. Here we have an annual species which does not cross either naturally or artificially with other species of the genus. Nevertheless it has given rise to hundreds of varieties including a wide range of color, form of flowers and habit of plant. On the other hand is *Lathyrus latifolius* of perennial character, of limited color range but with a larger number of flowers on the spike. Many years ago, as well as in later years, efforts were made to cross these species without definite result. The same seems to be true of attempts using other species. Now it is our hope that some one of the numerous species of *Lathyrus* will prove to be a key to unlock the riddle so that we may be able to cross *L. odoratus*, *L. latifolius*, *L. splendens*, *L. pubescens*, etc., with the result that we may have perennial peas in many colors as in the sweet pea.

In conclusion we may say that while species and varieties may be regarded as sterile when selfed, from the standpoint of the horticulturist no species or variety is really sterile until it can be shown that no cross is possible out of the multitude of forms available. Knowing that the flower is capable in itself of cross-fertilization there is always the lure to the plant breeder of some time making the fortunate discovery, if he tries long enough and persistently enough. These uncharted areas in the plant domain present a challenge for the future.

## STERILITIES ENCOUNTERED IN THE BREEDING OF PEONIES

A. P. SAUNDERS

*Hamilton College*

The work which I have been carrying on for the past ten years on peony species was undertaken from a purely practical point of view, in the effort to create desirable new types of garden peonies. What observations I have made on sterilities have been incidental. Hence the material I have to present is unavoidably somewhat scrappy and incomplete, though it covers a fairly wide range or will do so when the results of another year or two are added.

The slow germination of peony seed and the very gradual maturing of the plant make progress in such work a matter of considerable time. One must usually wait at least five years, and often more, to see the results of a given cross. However, in the time that has elapsed since I began the work certain tangible results have appeared and several rather curious incompatibilities have come to light.

It will be convenient to divide my observations under three heads: (1) shrubby species; (2) herbaceous species; (3) crosses between one of these groups and the other.

In the shrubby section three species are known, *Paeonia moutan*, the ordinary Chinese tree peony, *P. lutea*, a bright yellow-flowered species, and *P. Delavayi*, similar to *lutea* in foliage and general character but with the flowers stained red either at the base of the petals or more or less throughout. The type *P. moutan* does not seem to be anywhere in cultivation though it has been reported by travellers in the mountainous regions of China. On the other hand there are hundreds of named garden varieties. In general the plants set seed very well whenever the flower is perfect. Some of the very double forms are sterile through transformation of the carpels. Bees are very active in these flowers and it is likely that they inter-cross freely, though no direct observations are available. Pollen tests were made this year on six garden varieties selected at random, and they were all highly viable. Almost all my plants are regular and abundant seed setters.

Lemoine in France has crossed *P. lutea* with the garden forms of *P. moutan* and has put several remarkable hybrids into commerce—double yellow peonies. He reports that his successful crosses have always been those in which *lutea* was the female, and that all attempts to produce seed by the reciprocal cross were unavailing. My own records show 27 failures in the cross *lutea* male on *moutan* female, but also 5 seeds produced, although none of them germinated. There appears to be a one-way compatibility here which we shall find repeated later on with other peony species. I understand that these hybrids of Lemoine's have never set seed.

With respect to the cross between *moutan* and *Delavayi*, no data are available except my experiments of this year. As the seed is not yet matured I can only say that there are prospects of seed from this cross using *Delavayi* as female, though it is too soon to speak of the results of the reciprocal cross.

*Lutea* and *Delavayi* being evidently closely related species cross freely as one would expect in both directions. I got 21 good looking seeds from 7 crosses of *Delavayi* on *lutea* last year, and this year the prospects are that both that and the reciprocal cross will give good results.

Turning now to the group of herbaceous species, we have first of all the great race of garden peonies—Chinese peonies derived from *P. albiflora*, native to China. The number of named garden varieties in this group runs up into the thousands. Next in importance come those which have been developed out of the European species *P. officinalis*. There are two or three dozen named sorts in this group. Then comes the long list of other species, with most of which little apparently has been done, though from some of them a few improved forms have been developed. This list would include such species as *P. arietina*, *P. anomala*, *P. Broteri*, *P. Corsica*, *P. corallina*, *P. decora*, *P. tenuifolia*, etc., and then the more interesting and more recently discovered Caucasian species *P. Wittmanniana*, *P. macrophylla*, *P. Mlokosewitschi*, and the two dwarf growing Chinese species *P. Veitchi* and *P. Woodwardi*.

Most of my work has had to do with crosses between the garden forms of *P. albiflora* (*sinensis* as it is usually called) and *P. officinalis* and the pure species *P. macrophylla* and *P. Mlokosewitschi*.

So far as the garden varieties of Chinese peonies are concerned, there seems to be a very wide compatibility wherever the condition of the flower permits. In a vast number of crosses of one garden variety on another I have encountered nothing that suggests incompatibilities in this group.

Crosses of the varieties of *albiflora* on those of *officinalis* show a somewhat more varied behavior. Most of the double forms of *officinalis* are without stamens, and although the carpels are well developed these plants rarely set seed of themselves under ordinary garden conditions. This applies to the varieties *rubra plena*, *rosea plena*, *alba mutabilis*, *anemoneflora rosea*, and probably some others; these all set seed, though not abundantly, when hand pollenized with pollen from Chinese sorts. In the case of the single varieties of *officinalis* crosses may be effected in the reverse direction. These hybrids are distinguished by their very vigorous tall habit. I have 41 seedlings of this strain which have reached the blooming age. Of these 7 are *sinensis* on *officinalis*, and 34 *officinalis* on *sinensis*. Some of these have bloomed for the past three seasons, many for the past two. An examination of the pollen this year showed 25 out of the 41 completely sterile, or practically so, 9 feebly viable and only 7 possessing any noticeable vitality, and even then not much. One seed was obtained last year from one of these plants, but ordinarily they produce nothing but enormous empty seed pods. The phenomenon of parthenocarpy is rather widespread among peonies. I shall speak of it



again later on. The pollens of this group of hybrids approach a condition of uniform sterility; but even here those showing absolute sterility under the artificial conditions of the test are not more than 50 per cent.

I will now take up the crosses of *officinalis* and of *sinensis* with the pure species *macrophylla*. This latter is a very early blooming plant which goes out of bloom some time before the first *officinalis* sorts open. Hence my crosses between these two have all been made in the one direction. In the case of *sinensis* I am able to have pollen sent to me by growers in the south early enough to use on my plants of *macrophylla*. I am therefore able to cross in both directions *macrophylla* and *sinensis*.

Of *macrophylla-officinalis* hybrids I have had so far 15 that have bloomed. Every one of these has very strong pollen, and in 1925 four of them set seed, two quite freely. Seed has germinated in three out of these four cases, though not very well. Evidently there will be an  $F_2$  generation to work with here. I have this year, 1926, used the pollens of all or most of these 15 plants on blooms of *officinalis* but it is still too soon to say with what results.

The largest group of my hybrids were obtained by crossing *macrophylla* on varieties of *sinensis*. This cross rarely takes in the opposite direction. Hence we have here another case of a one-way compatibility, though the one-way-ness is not here absolutely exclusive. My records show, using *macrophylla* pollen on *sinensis*, 54 failed crosses against 1,500 seeds produced; but the other way 20 failures against 8 seeds which have yielded 6 plants. *P. macrophylla* sets seed of itself quite freely.

The hybrids of this strain show many of the characters of the male parent dominant in the first generation. The foliage and habit of the plant in a very large proportion of them could be mistaken for that of *macrophylla*, but never looks like the female parent. The general character of the flowers is also very much that of the male parent. Most of the crosses were made upon white or light pink varieties of *sinensis*, and the progeny are all white or pale pink. I have some hybrids from dark red varieties of *sinensis* but they have not yet bloomed.

I have this year examined the pollens of 178 of these hybrids. Those showing complete or nearly complete sterility numbered 133 or 75 per cent; in 36 or 20 per cent the pollen was somewhat viable, and in 9, or 5 per cent, there was strongly viable pollen. Whether the best of these pollens can be used for crossing back upon *sinensis* I may know by the end of the present season.

In spite of the fact that so many of these are sterile as to their pollen, some of them occasionally set seed. I had in 1924 four seeds, one each from four plants, out of perhaps 25 that bloomed. Of these seeds two have germinated. In 1925 I had seven seeds from six plants out of about 50 that bloomed; of these seven seeds only one has yet germinated. Curiously enough this occasional power to set seed does not seem to be connected with the viability of the pollen, for in only one case out of eight plants which have set seed and whose pollen has been tested was there any noticeable viability

in the pollen, and four of them showed the pollen completely sterile. I suppose it is rather likely that the formation of seed in these hybrids may be apogamous; but it is too soon yet to say.

We come now to the crosses made with and upon the species *P. Mlokosewitschi*. This plant is yellow flowered and has a general appearance very unlike that of any of the species heretofore considered. The stem is red, the leaves often reddish green and very dull, and with leaflets more rounded than in most species. This species has a highly developed incompatibility. My records show a long set of failures to get any cross-bred seeds using *sinensis* as the other parent. It is true that seed is occasionally obtained, as it is likely to be in all crossing of peonies, due to the fact that the flower often fertilizes itself before opening. But in every case where I have obtained seed from *sinensis* or from *Mlokosewitschi* after using the other species as pollen parent, the plants on maturing have shown the entire characters of the female parent. My records show 62 failed crosses of *Mlokosewitschi* on *sinensis*, and 34 failures with *sinensis* on *Mlokosewitschi*. It is to be noted that the latter species sets seed very freely to its own pollen. I have also used pollen of *Mlokosewitschi* on various forms of *officinalis*, and have a record during the past five years of 36 failures against 8 seeds, none of which germinated.

The species *P. Wittmanniana*, which is also yellow flowered, has been used by Lemoine as pollen parent in the production of those beautiful plants which are in commerce under the names Le Printemps, Mai fleuri, and Avant Garde. M. Lemoine has, he states, never been able to get anything by using pollen of *sinensis* on *Wittmanniana* as female. So there is apparently a one-way compatibility here somewhat like that which I have observed between *sinensis* and *macrophylla*.

Lemoine reports further that his *Wittmanniana* hybrids have always proved sterile, so far as the production of seed is concerned. My own experience confirms this at least regarding Le Printemps. My records show 45 unsuccessful attempts to fertilize it as well as half a dozen failures using the pollen of Le Printemps on *officinalis* varieties. This year I tested the pollen of all three of the Lemoine varieties. Their pollen is not absolutely sterile, and I have some experiments under way this year to see whether it can be successfully used in a back cross on *sinensis*.

The last group of crosses to be considered are those between the groups, shrubby and herbaceous. Here I have encountered only failures. Using pollen of tree peonies on *sinensis* I had 44 failures with 9 seedlings produced, all of them turning out to be pure *sinensis*. With the reciprocal cross 20 failures.

Pollen of *lutea* used on *sinensis* gave 93 failures with 19 seedlings produced, all pure *sinensis*. *Sinensis* pollen on *lutea* gave two failures.

Tree peony pollen on *officinalis* varieties gave 10 failures, no seed.

The obscure phenomena of partial or apparent fertilization are well illustrated in peonies. The ordinary herbaceous Chinese peonies rarely if ever give a developed seed pod without having fully fertilized seed in it. All the

*officinalis* section give as a rule when artificially fertilized a large seed pod. Sometimes this contains a few small but fertile seeds, but quite as often it is entirely empty. The hybrids between *sinensis* and *officinalis*, which have much the character of the *officinalis* forms, always form very large seed pods whether hand pollenized or not. These seed pods except for the one case mentioned earlier, have always been entirely empty. *P. macrophylla* almost always when pollenized develops an enormous seed pod; this may be filled with undeveloped bright red seeds which are soft between the fingers; or it may contain also a few larger dark blue seeds; and these in turn may yield to pressure between the fingers and disclose themselves as nothing but skin. Or, in favorable cases these blue seeds may be fertile. *Mlokoewitschi* behaves in the same way as *macrophylla*. *Lutea* under artificial fertilization very often gives immense seed pods filled with large glistening seeds, none of them fertile but all bursting open under a little pressure. The species *tenuifolia* behaves in just the same way, and when crossed with pollen of *sinensis* or *macrophylla* almost always gives abnormally large seed pods packed with seeds that are all soft.

I am now extending my observations over a larger range of species and hope within a very few years to be able to present a further and more complete report.

I wish to acknowledge the valuable assistance given me in making pollen tests by Miss Eleanor Meneely and Mr. Parke A. Dickey. All pollen tests were made in 5, 10 and 15 per cent cane sugar solutions containing about 1 per cent of agar.





# STERILITIES IN CARNATIONS WITH SPECIAL REFERENCE TO INTERSEXES\*

C. H. CONNORS

*New Jersey Agricultural Experiment Station*

(WITH PLATE 1)

The carnation, as commercially grown, is one of the horticultural plants commonly propagated by vegetative means, such as cuttings and layers. For this reason seed production is not important in the florist's carnation, except for the production of new varieties.

The carnation is assigned to *Dianthus caryophyllus*. It is probable that at least four, and probably more, species have entered into the genesis of this plant. It is highly heterozygous, like many of our horticultural plants.

The data from which the following conclusions have been drawn are the result of breeding work carried on at this station over a period of fifteen years, during which time about 10,000 seedlings from controlled cross- and close-pollinations were grown. The parents used at first were commercial sorts, and later our own seedlings were used for the crosses. There are here reported the progenies resulting from crosses among five commercial varieties.

## SUMMARY AND CONCLUSIONS

I. At least ten types and three sub-types of flowers are to be recognized in carnations with respect to the relative development of pistils and stamens. These may be described as follows:

1. Male, due to loss of styles and stigmas, the ovary being normal in size and form: observed only in singles.

2. Male, due to ovular pistillody: observed in singles, commercials and bull-heads.

3. Male, due to modified and probably non-functional stigmas: observed in singles, commercials and bull-heads.

4. Perfect hermaphrodite, retaining form of pistils and stamens throughout the blooming season: occurring in singles, commercials and bull-heads of relatively low petallage.

5. Fluctuating hermaphrodite, i. e., hermaphroditic for one period of the year, pistillate with rudimentary stamens (type 7-c) during another period, hermaphroditic during a third period: observed in singles, commercials and low petallage bull-heads.

6. Constantly fluctuating hermaphrodite, with a reduced number of stamens at best and with a loss and gain of stamens in successive flowers with no apparent correlation: observed in singles and commercials.

---

\*Paper No. 312 of the Journal Series, New Jersey Agricultural Experiment Stations, Department of Floriculture.

## 7. Pistillate:

- a. Stamens contabescent, i. e., present but anthers shrivelled, pollen shrivelled: observed in singles and commercials.
- b. Stamens present but small and impotent: observed in singles.
- c. Stamens rudimentary: observed in singles, commercials and bull-heads.

8. Pistillate, all stamens transformed into petals, e. g., excessive doubling: observed in bull-heads, although sometimes singles and commercials appear in which all the stamens are petaloid, the result of heredity.

9. Asexual, due to median proliferation: observed only in bull-heads.

10. Asexual, due to ovular pistillody and rudimentary stamens: observed in singles and commercials.

II. Flower type 1, 2, 7a, 7b, 9, 10 are of rare occurrence, and type 3 is relatively rare although in some progenies up to 25 per cent are found.

Types 4, 5, 6, 7c and 8 are common types in seed progenies.

III. A summary of the results from crossing and selfing is shown in the following table.

TABLE 1

COMPOSITE PROGENIES FROM CROSSES AND SELF-POLLINATIONS SUMMARIZED AND COMPARED, TOTALS AND PERCENTAGES OF THE THREE PREVAILING FLOWER TYPES BEING GIVEN, BASED ON COMPLETE POPULATIONS

MATING	♂ x ♂	♂ s. p.	♂ ♀ x ♂	♂ ♀ x ♂ ♀	♂ ♀ s. p.	♀ x ♂	♀ x ♂ ♀
Totals.....	122	262	30	31	109	56	59
?.....	6	7	16	14	3	0	9
F <sub>1</sub> .....	No....	65	110	11	12	57	33
	♂ P. C....	53.3	42.0	36.7	38.7	53.3	55.9
	No....	30	50	1	3	17	5
	♂ ♀ P. C....	24.6	19.1	3.3	9.7	15.6	8.5
F <sub>2</sub> .....	No....	19	91	2	2	30	12
	♀ P. C....	15.6	34.7	6.7	6.5	27.5	20.3
Totals.....	200	.....	35	71	.....	34	117
?.....	.....	.....	1	7	.....	.....	59
F <sub>1</sub> .....	No....	106	.....	15	21	.....	25
	♂ P. C....	53.0	.....	42.9	29.6	.....	21.4
	No....	44	.....	7	4	.....	3
	♂ ♀ P. C....	22.0	.....	20.0	5.6	.....	2.6
F <sub>2</sub> .....	No....	45	.....	11	39	.....	29
	♀ P. C....	22.5	.....	31.4	54.9	.....	24.8
Totals.....	.....	.....	.....	9	.....	.....	18
F <sub>1</sub> .....	No....	.....	.....	1	.....	.....	7
	♂ P. C....	.....	.....	11.1	.....	.....	38.9
	No....	.....	.....	1	.....	.....	.....
	♂ ♀ P. C....	.....	.....	11.1	.....	.....	.....
F <sub>2</sub> .....	No....	.....	.....	7	.....	.....	11
	♀ P. C....	.....	.....	77.8	.....	.....	61.1

A. Perfect hermaphrodite  $\times$  perfect hermaphrodite produced in the  $F_1$ , total 122, not recorded, 6, 65 perfect and 30 fluctuating hermaphroditic and 12 pistillate, 77.9 per cent hermaphroditic and 15.6 per cent pistillate, about 5 to 1.

Progenies from different parents behave differently, as 175f  $\times$  White Enchantress gave, total 39, 22 perfect and 4 fluctuating hermaphrodites, and 11 pistillate, a ratio of about 2 to 1. The other progenies behaved in a uniform manner, so there are at least 2 kinds of parents concerned with respect to the transmission of femaleness.

In the  $F_2$  selfed, two lines were studied, one from fluctuating parents and the other from perfect parents derived from a line in which the male progenitor tends to transmit femaleness in strong degree. In the first case the ratio is about  $2\frac{1}{2}$  hermaphrodites to 1 pistillate and in the second, about  $3\frac{1}{2}$  hermaphrodites to 1 pistillate. Both show a tendency to lose maleness in selfed lines, more evidenced in the first case but shown in the second, possibly, by the greater number of fluctuating hermaphrodites.

B. Perfect hermaphrodites selfed showed in the aggregate total of 262 (7 not recorded), 110 perfect and 50 fluctuating hermaphrodites and 91 pistillate, a strong tendency towards loss of maleness. Various individual parents vary, some producing more hermaphrodites and some fewer.

C. Fluctuating hermaphrodite  $\times$  perfect hermaphrodite gave, total 30 (not recorded 16), 12 more or less hermaphroditic and 2 pistillate. In the  $F_2$  (selfed), out of 35 (1 not recorded), 22 were hermaphroditic and 11 were pistillate.

D. Fluctuating hermaphrodite  $\times$  fluctuating hermaphrodite produced, total 31 (not recorded 14), 15 more or less hermaphroditic to 2 pistillate. In the  $F_2$  (selfed) were 25 hermaphrodites to 39 pistillate; and in the  $F_3$  (selfed) 2 hermaphrodites to 7 pistillates, the expected tendency toward femaleness being shown.

E. Fluctuating hermaphrodites selfed gave, total 99 (not recorded 3), 47 perfect hermaphrodites, 17 fluctuating hermaphrodites and 30 pistillate, a larger percentage of forms possessing maleness than from the perfect hermaphrodites selfed.

F. Pistillate  $\times$  perfect hermaphrodite produced, total 56, 18 perfect and 14 fluctuating hermaphrodites and 24 pistillate, about one-half being pistillate and approximately three-fourths more or less lacking maleness. In the  $F_2$ , total 34, the majority being from perfect hermaphrodites, 16 were perfect and 11 fluctuating hermaphrodites and 7 pistillate, about half being more or less lacking in maleness. The influence of the perfect hermaphroditic parent is apparent in the increased number of individuals possessing maleness.

G. Pistillate  $\times$  fluctuating hermaphrodite gave, total 59 (unrecorded 9), 33 perfect and 5 fluctuating hermaphrodites and 12 pistillate, the pistillate being about one-fourth of the total; in the  $F_2$  (selfed) were total 121 (unrecorded 59), 25 perfect and 3 fluctuating hermaphrodites and 29 pistillate; in the  $F_3$  (selfed) were 7 perfect hermaphrodites and 11 pistillate. The fluctu-

ating hermaphrodite appears to carry a stronger factor for maleness than does the perfect hermaphrodite, but there is a reduction in maleness in the selfed lines with this type also.

IV. Selfing either perfect hermaphrodite or fluctuating hermaphrodite tends to decrease maleness while crossing seems to maintain maleness.

V. Aberrant or monstrous forms are more likely to occur in selfed lines than in crosses.

VI. Environment affects sex manifestation during the period of the year when dull days occur in great number. The flowers whose primordia are formed during January and February (blooming during February, March and April) either lose maleness through contabescence of the stamens or the stamens are normal and plump pollen is formed which will not germinate under the conditions tried. Nutritional conditions are no doubt involved.

VII. Doubleness affects sex in the bull-heads or extreme doubles, when maleness and sometimes femaleness as well are lost, and a few cases of singles and commercials in which stamens have been transformed into petals. In another sense, maleness is vitiated by the division and increase in number of stamens with reduction in size of filaments and change in shape of the anthers.

VIII. Environment in affecting sex manifestations affects doubleness, for coincident with the loss of maleness or of male potency appears an increased doubling in the flowers of the commercial form.

IX. All three flower forms—single, commercial or half double and bull-head or extreme double—exhibit most of these flower types and types of intersexes.

X. The presence of intersexual forms is not of much importance in commercial breeding. Apparently, aberrant forms arise from any cross, but appear to be more common in crosses between related varieties or in selfed lines. Median proliferation and bull-heads can be avoided by crossing single and commercial rather than two varieties of the commercial form. In either case, about 50 per cent of the progeny will be commercials. Many of the aberrant forms can be eliminated as soon as the seedling plant becomes large enough to give some evidence of its character. If the inter-nodes are short, if the leaves are very broad and relatively short, the indications are that such individuals will be of the modified stigma type and hence of no commercial value. This can usually be detected in the cotyledons, which are much thicker, more rounded, often divided, in the undesirable forms.

XI. The carnation is in a dichogamous condition and appears, according to the evidence, to be tending toward a dioecious condition when allowed to self, i. e., close-fertilize.

#### EXPLANATION OF PLATE 1

Types of flowers in carnations.

1. The male flower.
2. A typical pistillate flower.
3. A typical hermaphrodite with stamens all dehiscent and stigmas not yet receptive.
4. Two flowers from a fluctuating hermaphrodite plant; flower at left pistillate; flower at right perfect.





CONNORS: STERILITIES IN CARNATIONS



# STERILITY ENCOUNTERED IN ROSE BREEDING

J. H. NICOLAS

*Research Department of The Conard-Pyle Company*

Sterility in roses is very frequent, although the pollen of most sterile seed-bearers is potent on other varieties, only two cases of sterility of both sexes having come to my knowledge, one being a double form of yellow *Rosa xanthina* and the other, Agnes, a hybrid of *R. Rugosa* and Persian Yellow of recent introduction. Several species are nearly sterile with their own pollen and entirely so with foreign pollen, while their pollen is potent on other varieties; such as *Rosa bella*, *Rosa alpina* and its hybrids, *Rosa lutea* and *Rosa centifolia*. Almost every moss-rose is sterile, the only exceptions I know being Blanche Moreau, Mme. Louis Leveque and Henri Martin, but the latter has so little moss to bequeath that nearly every one of its hybrids and even selfings are without moss. Blanche Moreau seedlings have so far been worthless and Mme. Louis Leveque reverts, no matter what pollen has been used, to the *centifolia* type, losing its moss altogether. In the days of the moss-rose popularity, a certain number of named varieties were introduced, but these were either bud mutations or merely pollen hybrids having only a suspicion of moss. Often their only claim to the name of moss was a glandular calyx emitting the peculiar scent of the true moss-rose.

Major Hurst of Cambridge University made a special study of the moss-rose and its sterility or fecundity. The moss-rose originated from bud mutations of the *R. centifolia* and all these bud mutations of first generation were sterile, but those of second generation (bud mutation from bud mutation) showed a certain fertility, but these were only lightly mossed. Quoting Hurst, "No records of the production of fertile seeds by either the typical *R. centifolia* or *R. muscosa* can be traced in botanical or horticultural literature. No fully formed and mature fruits have been observed on either form at Burbage (his old family estate) under normal conditions, during the last 70 years, though partly formed fruits containing no seeds have been frequently found." However, this sterility or rather infertility of the moss-rose is not always due to the involuted multiplication of the petals of the flower peculiar to both the typical *R. centifolia* and *R. muscosa* whose tightly balled petals and petaloid stamens seem to inhibit the natural development of pistils. The only fruits I ever obtained from Blanche Moreau and Mme. Louis Leveque were through artificial operation, when at an early age the petals, petaloid stamens, and stamens were carefully removed, and the pistils released and allowed to develop freely. Even at that the results were only one fruit to twenty failures, and the seedlings proved either worthless or identical to the mother. I tried again this summer on the mossiest of all the

moss-roses, an old variety known as English Pink Moss. At first, about half the operated blooms showed signs of impregnation, grew to a certain size and presently dried up. I understand that several hybridists lately have taken up again the moss-rose, but with what success I know not.

I mentioned Agnes as a patent example of sterility in *R. rugosa* hybrids. A close study of its organs showed misformed pistils and ovaries, the potency of its pollen, although pointing to sterility, is still undefined because of its paucity of stamens, which as in the moss-rose are covered and smothered by petals and petaloid stamens. I was occasionally able to free some of the anthers which seemed to release pollen, but which, however, brought no result, although applied to proved seed bearing varieties.

When the *R. rugosa* species became known, it was heralded as the fore-bearer of a new race of garden roses, and men like Gravereaux and Cochet took it up with enthusiasm, but the hybrids thus produced proved almost all sterile as to seed bearing, some at the first generation; others at the second, thus precluding the perpetuation, through a long descendance, of the commending features of the *rugosa* species, and to retain these dominant features in their entirety, or nearly so, the results so far obtained seem to indicate that the *R. rugosa* should be used as seed bearer; in the hybrids produced from *rugosa* pollen, the *rugosa* characters are so subdued as to be sometimes hardly recognized. Microscopical examination of the pollen of these three species (*centifolia*, *muscosa* and *rugosa*) is reported by Major Hurst as follows: "In *centifolia* and *muscosa*, 95 per cent of the grains were malformed, the chief difference being the presence in *R. muscosa* of a number of abnormally large grains showing signs of hypertrophy, and in both forms, degeneration of pollen was found in the early tetrad age. This result contrasts remarkably with the 99 per cent of well-formed grains found in *R. rugosa* and *R. arvensis* in all stages of development, but resembles rather the large percentage of malformed grains found in *R. Wichuraiana* and *R. laxa*."

The malformation of *R. Wichuraiana* pollen grains is probably the reason why, while we have so many *Wichuraiana* hybrids, I do not know of any issued from *Wichuraiana* pollen. From my own experience the pollen of *Wichuraiana* hybrids has invariably been sterile, yet the *Wichuraiana* type self-seeds very readily, practically every bloom being fertilized and the percentage of germination of these seeds is high.

A great many rose species hybrids are sterile after the first generation, and it was a great handicap to Dr. Van Fleet in his species hybridizing not to be able to carry on further generations.

Among the horticultural varieties of roses, many hybrids are sterile. The Bourbon strain at one time offered great promise, but the sterility of the hybrids discouraged the breeders and that strain has practically been abandoned. Souvenir de la Malmaison, one of those sterile Bourbons, gave me once a well developed selfing with three seeds, two of which germinated, only one surviving, the general character being Bourbon, but with enough changes in both foliage and bloom to surmise foreign pollen, although it might have



been a case of parthenogenesis. Since then I have again and again tried artificial pollination, but without success.

A case of near sterility worth noticing is American Pillar. It produces a profusion of fruits which in the fall and winter are very ornamental, but many hips are seedless; in the others, one or two seeds only, mostly empty, and a casual one only out of a large quantity of seeds will have a live germ.

The garden roses of today are so highly hybridized, many representing a succession of selected selfings, that a large percentage have run their course of fertility. Some are sterile with one pollen of proved potency and fertile with another. Although the pigmentation of two prospective parents may not have direct influence on fertility, yet I have observed that the more extreme the combinations the less successful I was in obtaining fertility. As an example Hawlmark Crimson, a blackish crimson, is sterile both ways when crossed with a deep yellow, and also is *R. Moyesii*.

Have the soil and original method of propagation a direct relation to the fertility or sterility of a plant? We have long noted here that grafted plants of *R. Hugonis*, for example, will profusely bear seeds, while plants grown from cuttings are very scant seed bearers, almost approaching sterility. Paul's Scarlet Climber as an own root plant may be considered as sterile, but a grafted plant will bear both self- and hand-pollinated seeds. I have also noted that plants of the same variety in different parts of the nursery have a different seed bearing capacity, although both receive the same amount of sunshine. As an instance, *R. bracteata* and *R. Altaica* at one location are practically sterile, while a short distance away, but in a different soil, nearly every bloom, either hand-or self-pollinated, sets fruit.

Climatic conditions may also have something to do: a rose hybridizer of experience in Canada writes that Mme. Caroline Testout and Gruss an Tep-litz are totally sterile for him, yet they notoriously are prolific seed bearers in other sections and in Europe.

How long does rose pollen retain its vitality? I do not know whether this question has ever been answered. I have obtained fertility by pollen preserved from two to three months in absorbent manila envelopes. P. Nabonnand of France told me that he used pollen a year old, and Pernet Ducher was successful with three-year-old pollen, both claiming that pollen to retain its potency must be in a container that will absorb moisture, and warned against using glass tubes.



## STERILITY AND INBREEDING IN BEETS

ERNEST REED

*Syracuse University*

The average sugar in the beet has not been changed since Mendelism was brought to light in 1890. The average sugar in the beet of the crop of Germany in 1882 was 13.1 per cent. The same figures may be used for the beet crop of the state of Michigan for 1921. There are on record occasional beets having tested 18, 20 and even 23 per cent. These individual tests, however, are of no genetical significance when one considers the variable factors involved. It is safe to say that the average sugar in the beet has not changed during its long commercial history.

This one characteristic of the beet is here chosen, not because of its commercial significance, but for the reason that plant breeders and geneticists have spared neither effort nor money in attempting to determine its heritable nature. Certain breeding methods have been and are now used in the production of the commercial strains of beets. Parent beets are selected on the basis of their sugar content and size. These selected beets are then isolated en masse for seed production. This method rests upon the assumption that sugar content is heritable. It is purely an assumption for we know nothing of the genetics of this characteristic in beets.

Pritchard (1) claims that there is no correlation between morphological characters and sugar content, and further, that sugar content is not inherited. There is a fallacy underlying the commercial beet breeding methods and the same may be said of Pritchard's method for they are fundamentally the same. In these cases the strains are produced by mass selection. The beet was first used as a foliage plant and selection was for color primarily. With such apparent characteristics desired, mass selection would account for the development of various color types. This method, however, has utterly failed in improving beets from the standpoint of sugar content.

As yet, we know nothing of the nature of the inheritance of sugar and since it is quite apparent, as Pritchard points out, that there is no correlation between morphological characters and sugar content, we have no characteristic to use in mass selection. The nature of the inheritance of sugar must be determined through inbreeding methods. The mass selection method is merely open hybridization in so far as sugar factors are concerned and this may readily account for the wide variability ranges of Pritchard and others.

The argument may well be urged that mass selection in beets maintains the stock at a high sugar value level. This may or may not be true. It is highly probable that the sugar value would not lower without mass selection. At least there is no data at hand which warrants the extensive expenditures involved in the mass selection methods.

In so far as has been determined experimentally the sugar content is effected only by hybridization with extreme types which are known to be very low in sugar content, such as the red garden beet, the yellow turnip beet or the half-sugar mangel wurtzel (4). The mass selection method is not required for the detection of these hybrid types, for they may be easily detected from apparent characteristics of color, shape, or size without the necessity of costly testing of sugar content.

### INBREEDING IN BEETS

Mass selection has been universally used in beet breeding for two reasons. It has been assumed that the beet stock will deteriorate if inbred. This assumption has no experimental status, and should receive little consideration until it has.

The second reason for mass selection lies in the difficulties involved in selfing. In surveying the history of beet breeding, one is impressed with the general sterility of beets, so much so that one might feel justified in saying that beets are self-sterile. The question, however, is far from settled. At the present, beet breeders in the western United States claim that they have no difficulty with sterility in beets, while European breeders find their beets self-sterile.

The writer has found beets to be normally self-sterile.\* The sterility is due to the protandrous character of the beet flower. Further, the self-sterility of the beet need not prevent inbreeding for ample seed may be secured by close-pollination.

### MATERIALS

The work presented here covers a period of ten years. Sugar beet seed was primarily used, but during the last four years parallel studies included the red table beet, the yellow turnip beet and the half-sugar mangel. Among the sugar beets were included every known commercial strain. It is needless to name and differentiate the many commercial sugar beet strains for they are all primarily the same in so far as one may determine. Types of any commercial variety occur in the other commercial varieties. There may be minor exceptions to this, as in the case of the strain known as "Dippe K," which may be distinguished by the color of its root epidermis.

### EXPERIMENTAL STUDIES

The primary object of the writer in this work was the genetical study of the beet. To this end methods were devised for the selfing of the beets but sterility was repeatedly encountered. Methods were then devised for insuring seed from close-pollination which amply serves the purpose for the genetical studies.

In the spring of 1917 eighty-four sugar beets were isolated by distance.

---

\* In this paper the types of fertilization are distinguished as by Shaw (2), "(1) self-fertilization, meaning that effected by pollen of the self-same flower, (2) close-fertilization, referring to that effected by the pollen of one flower upon any other flower of the same plant; cross fertilization, that effected between the flowers of any two plants."



No beet was within a half mile of any other flowering beet. Though these beets were carefully tended and flowered normally, producing abundant pollen, seventy-six of the plants yielded no seed. The other eight plants bore but two to ten seeds per plant.

The same methods of isolation were repeated in succeeding years with similar results. In 1920 eighty-four plants were put out, in 1921 one hundred sixty, and in 1922 three hundred. The majority of the plants produced no seed while no plant produced over twenty.

In 1921-24 beets were selected within a mass of Vilmorin stock. These plants were sacked with large paper flour sacks which covered the entire plant. The sacks were tied shut at the base of the stalks with cotton about the stalks to prevent the entrance of insects. There were ninety-six beets in the Vilmorin mass. The twenty-four sacked beets bore no seed while all of those open to cross-pollination bore heavily.

There was a possibility that if a beet could be induced to produce several seed stalks simultaneously, more flowers would be opening together and so the possibilities of seed would be increased by close-pollination. To this end four beets were sliced into longitudinal slices, two beets into two slices each, one into three; and one into four. The slices of each beet were planted together, the parts of one beet being isolated. The growth from these was poor and the sequence of flowering not changed, so there was no seed produced.

The seed yield of beets is known to be increased by removing the apical bud of each main stem, thus bringing on many lateral stems which would not otherwise have developed. The apical buds were removed from the main stalks of twenty of the isolated plants in 1922. Several hundred seeds were born on each of these plants. The same season twenty-four plants were cut back in this manner and hooded as well. Twelve were hooded tightly and twelve loosely. Insects were kept from entering those hooded tightly, while they had free access to those hooded loosely. The plants hooded tightly bore less than thirty seeds each, while those hooded loosely bore the heaviest yield of seed of any plants thus far self-pollinated. This method of isolating beets, cutting back the main stems and hooding the plant loosely insures a good yield of seed by close-pollination.

### THE BEET FLOWER AND PLANT

Darwin and others have pointed out that the beet flower is protandrous. The pollen is ready and gone from the stamens as much as three days before the stigma of the same flower are ready. This necessitates self-sterility in the beet.

The number of stalks vary greatly from one to as many as thirty. When the stalks are cut back numerous lateral stems grow out, greatly increasing the number of flowers. When such plants are hooded tightly and without insects within the hood there is no means for the transfer of pollen from flower to flower. Close-pollination then does not take place. When plants are hooded loosely insects transfer the pollen and close pollination is effected.

It has been pointed out that the paper sack hood aids in some manner, though it is not necessary for keeping out foreign pollen when the beets are isolated. Though no analysis of this phenomenon has been made the writer wishes to suggest that it may be due to the extra activity of the insects in the shade of the paper sack. Thrips may be more active about the plant in the shade than in the sunlight.

Shaw (3) has found that the beet flower is self-sterile when the pollen of a flower is saved and later applied to the receptive stigma. He also found that there was no sterility between flowers of the same plant but on different stems or stalks, thus suggesting a condition of gametic-sterility associated with physical proximity.

### DISCUSSION

Natural self-fertilization in beets is impossible because the pollen ripens and disperses before the stigma are mature. This element alone is sufficient to decrease the seed yield from the normal in inbred beets. In addition to this cause of self-fruitlessness, however, Shaw points out that the pollen of a flower will not function with the egg-cell of the same flower. He saved pollen from certain flowers and placed it on the stigma when the latter were mature and no seed formation ensued. This indicates a possible gametic incompatibility within the flower.

Shaw further shows that this gametic incompatibility diminishes as the distance between the flowers increases; that close-fertilization is possible between flowers of different stems of the same plant.

The experiments cited in this paper do not include studies which might verify the hypothesis of gametic incompatibility within a flower. But the studies do show that the close-fertilization gives a striking increase of seed yield with an increase in lateral branches which follows the severance of the apical buds.

During the progress of this work pollen of five flowers was saved and later applied to the mature stigma. Two seeds resulted from this self-fertilization. This is such a meager offering on this phase of the work that it hardly deserves mention. It does, however, lead the writer to the possible conclusion that the lack of seed in such cases is due to imperfect technique rather than to gametic incompatibility. Relative to the effects of hooding on seed production it should be pointed out that the hooding no doubt effects the physiological processes of the plant. This suggests that physiological incompatibility may be operating in sugar beets; that physiological processes which normally restrict fertilization may be effected by the hooding, which, no doubt, modifies light, heat and humidity factors (3).

Though there is at least one cause of self-fruitlessness in beets, close-fertilization may be relied upon to insure sufficient inbred seed for genetical studies.

### SUMMARY

The self-fruitlessness is due to the protandrous character of the beet flower.

Beets which flower in isolation bear a very small amount of seed, in which cases the fertilization is close and probably due to insects.

Beets hooded tightly without insects included within the hood bear no seed.

Beets hooded loosely, allowing insects to enter, though isolated by distance from other flowering beets, bear seed. The amount of seed is greatly increased by cutting back the apical buds of the main stems, thus bringing on many lateral stems.

### LITERATURE CITED

1. Pritchard, Frederick J. Some recent investigations in sugar beet breeding. *Bot. Gaz.* 62: 425-465. 1916.
2. Shaw, Harry B. Self, close and cross-fertilization of beets. *Mem. The New York Bot. Gard.* 6: 149-152. 1916.
3. Stout, A. B. The physiology of incompatibilities. *Am. Jour. Bot.* 10: 459-461. 1923.
4. Vilmorin, M. J. L. *L'hérédité chez la Betterave cultivée.* 1923.





## CORRELATION AND CYCLIC STERILITY IN CLEOME

A. E. MURNEEK  
*University of Missouri*

While many types of sterility and unfruitfulness in higher plants are quite certainly due to inherited genetic causes, others are clearly the result of changes in environment external to the germinal tissues. Considerable interest has recently been centered on such manifestations of sterility of evidently a physiological nature that occupy an intermediate position between a state of true fertility and one of complete sterility. "Partial sterility," "pseudo-sterility" and "cyclic sterility" are some of the terms used to express the nature of this condition. Though by no means synonymous in their application, the terminology is fairly indicative of the general situation.

This "twilight zone," as it were, of sexual impotence appears to be a particularly tempting field of special inquiry. The reasons are rather evident. Under the conditions of partial or incomplete sterility probably are exhibited various stages of transition of sexuality. The direction of change may be either toward an ever increasing sterility or else toward a higher state of fertility.

The situation may be analyzed with two major points of interest in mind. First, some of these types of sterility are of considerable significance in studies of the evolutionary development of a species or a variety. So, too, they are of profound theoretical and practical value in connection with plant improvement and propagation. A second point of interest is the manifestation of sexuality and fruitfulness of various degrees of intensity during the ontogeny of a plant. This condition seems to be especially favorable for experimental study of the physiological basis of sterility.

Two very characteristic instances of cyclic manifestations of sterility or intermittent fruit production have been carefully observed and discussed by Stout. In several strains of two species of *Brassica* (*B. pekinensis* and *B. chinensis*) sterility may be due to at least three causes (Stout, 1): (a) Impotence, manifesting itself either in arrested development or abortion of flowers. (b) Proliferation, due apparently to excessive vegetative vigor. (c) Physiological incompatibility, conditioned most likely by the internal state of nutrition. All these types of sterility evidently are associated with alternations in vegetative and reproductive vigor of the plant. Their appearance is distinctly cyclic and coordinated among the different branches. Hence the phenomenon is very suggestive of having not only a physiological basis but also a physiological coordination.

A distinctly different kind of cyclic sterility is illustrated by *Cleome spinosa* (Stout, 2). The various grades and degrees of sex expression in the

hermaphroditic flowers of this vigorous annual are apparently conditioned by internal physiological changes resulting in intermittent fruitfulness. This species is sterile only as far as it is fruitless. Unlike the cyclic manifestations of sterility in *Brassica*, alternations in sexuality in *Cleome* have been correlated neither with changes in external environment nor with the general vegetative vigor. According to Stout, cyclic fruitfulness in this plant is most likely due to internal biogenetic regulation and is possibly directed by specific stimulating or inhibiting influences.

The present paper is the result of the writer's interest in *Cleome* as a material and in analysis of cyclic sterility as an avenue of approach to studies of the physiological basis of certain forms of sterility in horticultural plants. The work is still in the inception stage. The information so far obtained, however, seems to be of sufficient interest and value to be related here in the form of a summary. The detailed data will be published elsewhere.

### EXPERIMENTAL RESULTS

The experiments upon which this report is based consisted of several series of plants, grown in pot cultures of 12 liter capacity under a standard greenhouse environment. All of the specimens were raised during the late winter and early spring months from seed collected at random. To obviate any disturbances that might result from secondary vegetative growth, the plants were trained to a uniaxial type of development. This is easily accomplished by the prompt removal of all axillary shoots.

An attempt was made to answer two questions: (a) What is the general influence of nutrition on cyclic manifestation of fruitfulness? (b) What, if any, are the effects of correlation between the developing fruit and sex expression of the succeeding flowers? The conclusions are drawn from observation of more than 10,000 flowers, the development and behavior of 8,638 of which have been carefully recorded. Hence the results are thought to be quite typical.

Nitrogen being one of the most important and often a limiting factor in nutrition, groups of plants were grown on two levels of nitrogen supply. Some were raised in pure quartz sand on a six-salt nutrient solution devoid of nitrogen. Others were grown in very rich garden compost, which was watered at frequent intervals with a weak solution of  $\text{NaNO}_3$  and  $\text{KNO}_3$ . Thus plants of two somewhat extreme types of growth were secured: one low in nitrogen and weakly vegetative, the others high in nitrogen and very vigorous.

Having reached a certain stage of development, plants of both series changed from a vegetative to a reproductive state. Those in sand cultures preceded the soil series by approximately 30 days. All open flowers were pollinated daily by hand. It is thought that a high degree of fertilization was accomplished by this practice.

In every case reproduction in the vegetative plants began with highly functional hermaphroditic flowers of the usual size for *Cleome*. The first

7-50 flowers exhibited normal stamens and pistils. As soon as the fruit pods began to develop, this cycle was followed either by a single or else a short cycle of staminate flowers, next a group of complete flowers, again a few staminate, etc. The rather typical average alternation was as follows:

♂ ♀      ♂      ♂ ♀      ♂      ♂ ♀      ♂      ♂ ♀      ♂  
 21 — 6 — 6 — 2 — 4 — 1 — 3 — 1....

Most of the flowers in this group were hermaphrodites. Intermittent production of fruit was due largely, if not entirely, to cyclic reduction in the development of pistils. In a few instances abortion of stamens was noted. Such more or less rudimentary male organs most likely produced various amounts of non-viable pollen (Stout, 2). This condition was not determined because of lack of time. The alternating cycles are spoken of as consisting of male and hermaphroditic flowers, since stamens always produced some pollen. Frequently the alternating groups were of distinctly greater amplitude than typified above, permitting the recognition of 13-14 large cycles during the production of some 300-350 flowers. In spite of the excessive supply of nitrogen, this series behaved very much like plants normally grown outdoors.

When *Cleome* was developed without nitrogen, as in the present sand cultures, sterility was still of a cyclic nature, but somewhat altered in intensity of expression of either sex. In approximately 10 per cent of cases the first cycle consisted of staminate flowers, which naturally were non-fruitful. Otherwise the cycles began as usual with hermaphroditic flowers and changed to male, to hermaphrodite, to male, etc. The average sequence was of the following order:

♂ ♀      ♂      ♂ ♀      ♂      ♂ ♀      ♂  
 4 — 10 — 4 — 4 — 2 — 3....

It is evident that staminate flowers predominated in this series. Moreover, the last flowers on these plants were either males or else highly reduced females. In fact, the only conspicuous effect produced by lack of nitrogen was an increase in maleness. The distinctly cyclic production of fruit was not altered to any marked degree.

When 30-35 blossoms had been formed on these plants further flowering ceased. Any buds that were still produced turned yellow and abscised. These, of course, were the unmistakable signs of waning vigor—a consequence of the developing fruit. For as soon as the pods had matured and released their contents of seeds, normal opening of buds recommenced, provided the individual had not become too far exhausted. This consequently leads one to the question of correlation.

In a recent study of the effects of reproduction on vegetative development in the tomato (*Lycopersicon esculentum*) the writer has demonstrated (Murneek, 3, 4) that during the period of fruit development not only vegetative growth is strikingly retarded, but that there is also a marked disturbance in the normal performance of the floral organs. A careful measurement of all specimens in the several groups of *Cleome*, including those described



forthwith, showed practically no effect of fruit production on vegetative extension of plants in the soil series, but an appreciable effect in those of the sand cultures. Evidently under strictly uniaxial conditions of growth with an ample supply of soil nutrients, all the activities in this species, when once initiated, are confined to reproduction. There is, however, a very profound effect of the developing fruit on morphogenesis of sexual organs, as the following experiments will show.

From representative specimens of both types of plants the first fruit was removed when about half grown, as judged by size. Under this treatment recovery in the nitrogen low group, from the usual inhibition due to fruiting, was rapid, if the pods had not attained too high a stage of maturity. When more or less mature pods were removed, recovery was slow and of a low degree. A few plants had become so far exhausted by the maturing of the first crop of pods that no recovery was obtained. Such individuals presumably had utilized all of the available or bound nitrogen, as indicated by abscission of almost every leaf. This condition resembles closely a similar response by the tomato when grown without an external supply of nitrogen.

Similarly, a periodic removal of immature pods from nitrogen high plants was accompanied by a shortening of the cycles of staminate flowers. In some instances this resulted in a complete change from intermittent to continuous production of hermaphroditic flowers, which were highly functional both as males and females. This then was very suggestive of the real nature and cause of cyclic sterility or periodic fruitfulness in *Cleome spinosa*.

Naturally one more step was required to complete the evidence. Hence from other groups of both nitrogen low and nitrogen high plants all pistils were cut daily, or as soon as formed. Those of the nitrogen low series responded to this treatment by the abbreviation of the first cycle of staminate flowers and a reduction of the second and third cycles of males to a single, rarely two blossoms. Following this an uninterrupted formation of hermaphroditic flowers was initiated, which did not cease till the plants showed signs of complete devitalization. In many specimens of nitrogen low plants under this treatment but one short cycle of male flowers was formed, all the rest being perfect.

A very similar response was exhibited by nitrogen high plants as a result of the prompt removal of the pistils. A rapid change from a condition of cyclic sterility to one of complete fruitfulness could be observed. Soon after initiation of depistillation a continuous succession of perfect flowers was produced without the slightest manifestation of the characteristic periodic alternation to maleness. Moreover, in fully 50 per cent of plants in this group complete hermaphroditism and hence fruitfulness was manifested from the very beginning, due to the daily removal of pistils. On these specimens not a single staminate flower was produced throughout their lifetime. Many of these plants were permitted to form over 300 flowers—all hermaphrodites. When fruit was allowed to develop the usual cyclic condition of sterility was readily initiated.



To make it more certain that variations in sex expression in *Cleome* is not of genetic but of purely nutritional or correlative character, a certain number of plants of the soil or nitrogen high series were trained to a diaxial type of growth. Such two-stemmed plants are easily secured by an early removal of all axillary growth excepting at two desired points. The main stem is cut off above these side shoots. One of the branches on each plant was left undisturbed, producing fruit periodically. Pistils were removed from the second in a manner detailed above. The results in all cases were as if two individual plants had been employed for the experiment. The depistillated half of each plant exhibited the characteristic inhibition of staminate cycles. After a certain time only perfect flowers were formed on the treated branch. During the same period the normal or control branch showed the usual intermittent alternation in sexuality and fruitfulness.

### DISCUSSION

Cyclic sterility in *Cleome spinosa*, as exhibited by intermittent production of fruit, is not due to impotence of both sexes. Even under very extreme conditions of nutrition and growth no flowers were secured in which both sexes were completely non-functional (2). Neither is physiological incompatibility a factor to be considered, as the careful investigations of Stout have shown. The condition is caused largely, if not entirely, by a conspicuous cyclic suppression of pistils of the otherwise perfect flowers. They remain functional only as males during certain recurring periods. Naturally no fruit is then formed.

It is evident that the phenomenon can not be due to inherited genetic causes, for how could we then explain, except by the assumption of some highly hypothetical mechanism, the complete reversal from a condition of cyclic sterility to that of continuous fruitfulness, as in the present experiments. Neither would it be possible to account genetically for the non-interrupted hermaphroditism in one-half of a plant, while the other half at the same time was exhibiting characteristic periodic changes in morphogenesis of sex organs.

There seems to be hardly room for doubt that intersexuality of the type manifested by *Cleome* has a strictly physiological basis, an important phase of which is the general state of nutrition of the plant. It was shown, for instance, that nitrogen may play a conspicuous rôle in this connection. Under restricted nitrogen nutrition sex expression was predominately male, while an excessive supply of nitrogen led to increased femaleness. This, however, has been repeatedly demonstrated with many other especially dioecious plants (Atkinson, 5; Schaffner, 6, 7; Gardner, 8). Moreover, such a conception leads to a logical explanation of the frequently observed reversals of sex late in the season in strictly pistillate plants (Schaffner, 9, 10; Rosa, 11). End-season alternations in sexuality are undoubtedly connected with the waning vigor of a plant, which in turn is associated with a low nitro-

gen supply. Of course, other indispensable factors of environment may play a similar rôle.

One should remember that in an annual plant with a determinate growth, or one trained to such form of development, the whole energy of the organism is usually confined to reproduction, once it is initiated. A most important feature of this condition is the nutrition of the gametophytes, the gametes, but especially the developing embryos and their accessory tissues. The demand to which the sporophyte is exposed by the support of a large crop of seeds and fruit can hardly be over-emphasized (4). Under conditions of comparatively limited absorption of soil nutrients or restricted organic synthesis, the vegetative parts of a plant may become totally exhausted due to the excessive drain by the embryos, leading to eventual death of the whole organism (Korschelt, 12). This would be particularly true of a polycarpic plant like *Cleome*.

Hypothetical as it may appear, still it is conceivable that "there are two distinct types of metabolism, one of which is male-determining, the other female-determining," as postulated by Geddes and Thomson (13) and recently emphasized by Brambell (14). In *Cleome*, however, this would necessitate the assumption of an oscillating alternation in special nutrition of the female organs and a more or less uninterrupted nutrition of the staminate parts of the flowers. Moreover, such a condition would have to be of the nature of smaller special cycles of nutrition superimposed upon and operating within the general larger developmental cycle of the sporophyte. If this is at all probable, it still would call for a fundamental causal mechanism of initiation of such recurring cycles of special nutrition.

The most important fact brought forth by the present experimental analysis of sexuality in *Cleome* is the striking effect of correlation between the developing fruit and sex expression of the succeeding flowers. If the recurring cycles of staminate flowers were due to changes in the general nutrition of the plant, then a drastic shifting of the level of nutrition, particularly nitrogenous, should lead to their disarrangement. This clearly was not the case. It is possible, however, that because of the practice of artificial pollination an exceedingly high degree of fertilization and fruitfulness was maintained in both nitrogen low and nitrogen high plants. The capacity of the organs of absorption and synthesis may thus have been continuously overtaxed, due to the excessive requirements for elaborated substances by the large crop of fruit. Hence in spite of a very favorable external condition of nutrition, as when *Cleome* was grown in rich soil, the internal supply of organic substances would still be constantly at a minimum. Assuming this to be the case, what, then, is the mechanism by means of which the developing embryos are able to monopolize most of the incoming or elaborated food supply?

In this connection the theory proposed by Child (15) is worth considering. Production of gametes may be interpreted as a sign of senescence in a plant, associated with a comparatively low rate of metabolism. Fertiliza-

tion or gametic union, on the contrary, is the beginning of rejuvenescence, initiating high localized levels of metabolism and maintaining them throughout the time of embryo development and their intimate contact with the mother sporophyte. Once a comparatively large number of such metabolic gradients are initiated, as in cases of excessive fecundity, it may lead to diverting of particular substances to the embryos or fruit and consequent disturbance in floral morphogenesis. The pistillate part of the flower, because of its relatively higher level of nutrition, would be the first to suffer in this regard. It should be noted that when the fruit is not allowed to come to maturity, no retardation in the development of pistils is observed. Consequently, a removal of the fruit pods during early stages of growth leads to prompt recovery from all conditions of sexual impotence. One must emphasize that a correlation of this nature is quite independent of the external environment. It is strictly of the character of internal physiological readjustment.

Finally, stimulating and inhibiting substances, specific enzymes or hormones, may also be instrumental in bringing about these effects. They may operate on the general metabolism of a plant as regulating agents. There is ample proof that such catalytic or inhibitory substances are not only in existence in the animal body, but that they are profoundly effective in the metabolism of certain organs during particular stages of development (Starling, 16; Bayliss, 17). The conspicuous results of this control are expressed in the development and function of various parts of the body. Such a system of secretions, being endogenous, may be initiated and may come into operation at particular stages of development of the sex organs, but especially during the growth of the embryo.

Whatever the detailed mechanism may eventually be by means of which sexual changes and periodic fruitfulness are brought about in *Cleome*, this much is clear at present: (a) The basic cause or causes are of a physiological nature. (b) Nutrition has a considerable effect in altering the sexual states. (c) A distinct correlation exists between fruit and sex expression. When fruit is not permitted to develop cyclic sterility will not occur.

#### LITERATURE CITED

1. Stout, A. B. Cyclic manifestations of sterility in *Brassica pekinensis* and *B. chinensis*. Bot. Gaz. **73**: 110-132. 1922.
2. Stout, A. B. Alternation of sexes and intermittent production of fruit in the spider flower (*Cleome spinosa*). Am. Jour. Bot. **10**: 57-66. 1923.
3. Murneek, A. E. Correlation and cyclic growth in plants. Bot. Gaz. **79**: 329-333. 1925.
4. Murneek, A. E. Effects of correlation between vegetative and reproductive functions in the tomato. Pl. Phys. **1**: 3-56. 1926.
5. Atkinson, G. F. Experiments on the morphology of *Arisaema triphyllum*. Bot. Gaz. **25**: 114. 1898.
6. Schaffner, J. H. Control of sexual state in *Arisaema triphyllum* and *Arisaema dracontium*. Am. Jour. Bot. **9**: 72-78. 1922.
7. Schaffner, J. H. The influence of the substratum on the percentage of sex reversal in winter-grown hemp. Ohio Jour. Sci. **25**: 172-276. 1925.
8. Gardner, V. R. Studies in nutrition of the strawberry. Mo. Agr. Exp. Sta. Res. Bul. **57**. 1923.
9. Schaffner, J. H. Influence of environment on sexual expression in hemp. Bot. Gaz. **71**: 197-219. 1921.
10. Schaffner, J. H. Sex reversal in the Japanese hop. Bul. Torr. Bot. Club **50**: 73-79. 1923.

11. Rosa, J. T. Sex expression in spinach. *Hilgardia* 1: 259-274. 1925.
12. Korschelt, E. Lebensdauer, Altern und Tod. Jena, 1924.
13. Geddes, P., and Thomson, J. A. Evolution of Sex. New York, 1911.
14. Brambell, F. W. R. Sex reversal and intersexuality. *Jour. Roy. Mic. Soc.* 1923: 395-408.
15. Child, C. M. Senescence and Rejuvenescence, Chicago, 1915.
16. Starling, E. H. The chemical coordination of the activities of the body. *Sci. Prog.* 1: 557-568. 1907.
17. Bayliss, W. M. Principles of General Physiology. Ch. 24. London, 1924.



# EFFECT OF LIGHT, CARBON DIOXIDE AND TEMPERATURE ON FLOWER AND FRUIT PRODUCTION

JOHN M. ARTHUR and JOHN D. GUTHRIE

*Boyce Thompson Institute for Plant Research*

(WITH PLATES 2-3)

This paper reports two series of experiments—first, a series of plants grown during 1925 at a temperature of 26° centigrade and, second, a series grown during 1926 at a temperature of 20° centigrade. Both series of plants were grown under the following four conditions:

1. Greenhouse control. Ordinary greenhouse conditions during March, April and May, but with temperature and humidity controlled.

2. Greenhouse 1. A similar greenhouse but with 6 hours additional light, 12 midnight to 6 A. M., from a gantry crane carrying 48 1000-watt gas-filled lamps.

3. Greenhouse 2. Same additional illumination as Greenhouse 1 except from 6 P. M. to midnight. This house received additional carbon dioxide gas, delivered to the house through a flow meter from steel cylinders. An attempt was made to hold the concentration of gas at about .3 per cent, although this was found to be very difficult when the greenhouse vents were open.

4. Constant Light room. Temperature and humidity controlled and with increased carbon dioxide concentration. Plants grown in this room received only artificial illumination. The illumination consisted of 25 1500-watt gas-filled lamps and 4 mercury vapor tubes.

Lantern slides were shown, giving briefly some of the results on flowering and fruiting as follows:

Several grains such as oats, spring wheat and barley were found to grow well and yield well even when grown continuously at a relatively high temperature (26° C.) if given additional light and carbon dioxide, as in Greenhouse 2. With additional light only, Greenhouse 1, fewer heads were formed and the plants were not so tall. Plants in Greenhouses 1 and 2 flowered and fruited much earlier than controls. Ripening heads were formed in both Greenhouse 1 and 2 in 45 days from seed, at a time when heads were not yet visible on the control plants. When the same grains were grown at a lower temperature, 20° C., the difference between plants grown under the different conditions was less marked but plants receiving additional light and carbon dioxide matured earlier than controls. Walster (1) found that barley grown at a high temperature with a high nitrate supply gave excessive vegetation and little culm formation. With additional light and carbon dioxide it was found that barley grows well and yields well even at a high temperature and with abundant nitrogen supply.

Potatoes grown at 26° C. produced very weak stems, few flowers, and only small tubers, even with additional carbon dioxide and light; while those grown at 20° C. produced strong stems, large tubers and flowered especially well in Greenhouse 2 with additional light and gas. The potato is definitely limited to a low temperature and is not able to utilize additional light and carbon dioxide at high temperatures.

As reported by Garner and Allard (2) length of day was found to have a marked effect on the flowering and fruiting of several plants. Several species of plants were grown under artificial illumination in the constant light room at different day lengths as follows: 5, 7, 12, 17, 19 and 24 hour exposures to light in each 24-hour period. Lettuce and radish flowered on day lengths greater than 12 hours. Buckwheat flowered on all day lengths from 5 to 24 hours. Salvia flowered on day lengths from 5 to 12 hours only. On longer day lengths salvia remained vegetative except where buds were already formed on the plants when they were placed under the controlled conditions. In no case, however, did salvia buds open on a 24-hour day during these experiments. Tomato plants were greatly injured by exposure to the longer day lengths. These plants died away completely on a 24-hour day. They withstood successfully a 19-hour day although their leaves were greatly injured. They set fruit only on 12, 17 and 19-hour days. Ragweeds (*Ambrosia artemisifolia*) were found to flower on the short days of spring in the control greenhouse but did not flower in either Greenhouse 1 or 2 where they received 6 hours additional light in each 24-hour period. The above day length considerations were found to be independent of temperature.

Sweet peas, petunia and snapdragon all flowered profusely on a 24-hour artificial day in the constant light room. In general this flowering was not greatly affected by temperature changes between 20 and 26° C., but sweet peas grew and flowered most luxuriantly at the lower temperature.

Red clover was grown from seed to flower in 38 days in Greenhouses 1 and 2, and in the constant light room, on 18 and 24-hour days respectively. Additional concentrations of carbon dioxide increased both the height of the plants and amount of flowering.

#### LITERATURE CITED

1. Walster, H. L. Formative effect of high and low temperatures upon growth of barley: A chemical correlation. Bot. Gaz. 69: 97-126. 1920.
2. Garner, W. W., and Allard, H. A. Effect of the relative length of day and night and other factors of the environment on growth and reproduction in plants. Jour. Agr. Res. 18: 553-605. 1920.

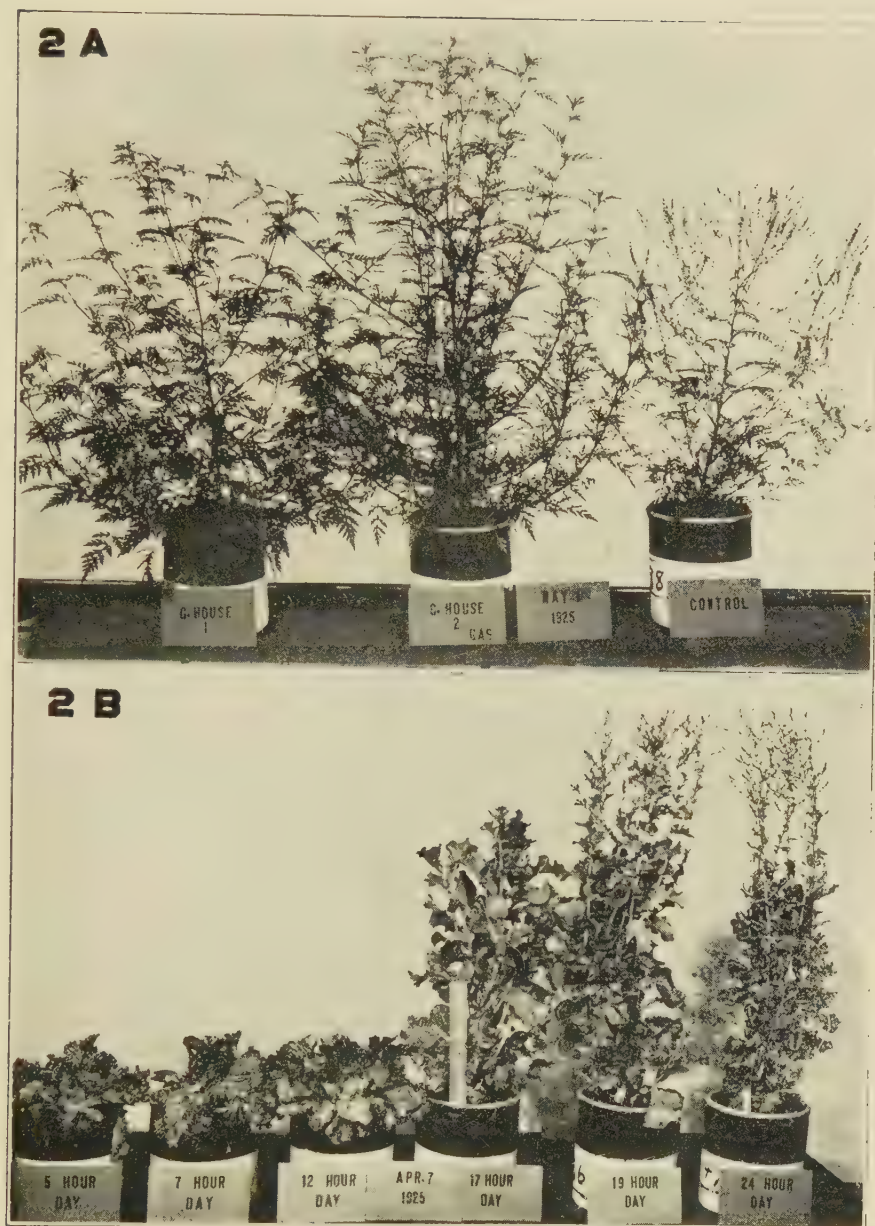


1A. Red Clover. Greenhouse 1, daylight plus 6 hours of artificial light. Greenhouse 2, same light plus additional carbon dioxide. 24 hour day plants grown under all artificial light. Control plants ordinary daylight of spring. Plants 69 days old from seed.

1. Barley. Grown in Greenhouse. 1, 2 and control with same illumination and carbon dioxide concentration as clover shown in Plate 3, 1A. Plants 41 days old from seed.







2A. Ragweed. A short day plant showing flowering habit on the short days of spring in control greenhouse and a tendency to remain vegetative with additional illumination in Greenhouses 1 and 2.

2B. Lettuce. Grown in constant light room on 5, 7, 12, 17, 19 and 24 hours of artificial light. A long day plant.



# PARTIAL STERILITY IN HYBRIDS OF SORGHUM AND JOHNSON GRASS

H. N. VINALL

*Bureau of Plant Industry, U. S. Department of Agriculture*

(WITH PLATE 4)

The late Dr. Charles V. Piper in Vol. 28, pp. 25-44 of the Proceedings of the Biological Society of Washington has the following to say regarding the botanical relationships of sorghum and Johnson grass:

"Johnson grass, *Andropogon halepensis* (L.) Brot., and sorghum, *A. sorghum* (L.) Brot., are nearly always treated as distinct species in botanical works. Along with this botanical treatment, however, the statement is frequently made that the latter is believed to be derived from the former under cultivation. This conclusion is usually accredited to Hackel but without due consideration to what that botanist actually wrote. It is true Hackel considered that there was but one botanical species involved, namely, *Andropogon sorghum*, but consisting of two subspecies, *A. sorghum halepensis*, wild perennial or rarely annual plants with the spikelets readily deciduous at maturity, and *A. sorghum sativus* mostly cultivated, annual or in the tropics sometimes perennial plants, with the spikelets persistent at maturity. While Hackel regarded the cultivated plants as having been derived by cultivation from *A. sorghum halepensis*, he expressly writes that he does not believe that the wild varieties with rootstocks were at all concerned with the cultivated sorghums, but that the latter originated from such wild varieties as *effusus*, *virgatus*, and *aethiopicus*, all of which are devoid of rootstocks.

"A more satisfactory treatment of the plants in question is to consider them two distinct species—*Andropogon halepensis*, perennial plants possessing rootstocks, and *Andropogon sorghum*, annual plants (perennating in frostless regions) without rootstocks. The facts of distribution as well as those concerned with the origin of the cultivated sorghums are consistent with this treatment."

Another binomial applied to Johnson grass by some writers is *Holcus halepensis* L. Dr. O. Stapf, in his consideration of the sorghums, published as late as 1917, in *Flora of Tropical Africa*, Vol. 9, part 1, pp. 104-154, follows Persoon and makes the genus name *Sorghum*. Thus, according to Stapf, Johnson grass is *Sorghum halepense*, Pers., and the sorghum groups are given various species classification, such as *Sorghum durra*, *S. caudatum*, *S. cerneum*, etc. Dr. Piper, in the revised edition of his book "Forage Plants and Their Culture," accepted the decision of Stapf as to the genus and refers to Johnson grass as *Sorghum halepense* Pers., but did not follow the latter author in his treatment of the annual sorghums, preferring to place all forms

in one species, thus: *Sorghum sorghum*, and consider the subdivisions of this group as varieties.

It is now rather generally conceded that Johnson grass and the cultivated sorghums deserve separate species classification, but whether we admit this or not the evidence points strongly to a much closer relationship between the annual grass sorghums, such as Sudan grass (*Sorghum sudanensis*), Tunis grass (*S. virgatus*), and Tabucki grass (*S. verticilliflorus*), and the cultivated sorghums, than between these sorghums and Johnson grass. In the first place, Johnson grass possesses rhizomes or underground stems, which make it perennial except in northern latitudes; secondly, its natural distribution in Africa, undoubtedly the birthplace of the sorghums, does not coincide so fully with the distribution of the larger forms of sorghum as does the natural distribution of the annual grass sorghums; and finally, these wild grass sorghums cross naturally and easily in the field with the larger cultivated varieties, while very few natural crosses of sorghum and Johnson grass have been discovered in our southern states where the two plants have grown side by side for years and artificial crosses are made with difficulty.

In studying the relationship of sorghum and Johnson grass numerous cross-pollinations of these two species were made in the autumns of 1912 and 1913, and only two were successful. The 1912 crosses were made in the field, those of 1913 in the greenhouse, and although several of the more important sorghum varieties were used as the pistillate parent and the pollen parent, the successful cross in both years was obtained with Black Amber sorgo as the pistillate parent. No success was attained either year in the fertilization of Johnson grass flowers with sorghum pollen.

Other crosses were made in the greenhouse during March, 1914, and again success was attained only with Dakota Amber, a dwarf strain of Black Amber sorgo. The seed resulting from the application of Johnson grass pollen to the emasculated flower on this variety of sorgo was planted in the greenhouse and the panicle of the resulting  $F_1$  plant was photographed to show the partial sterility existing in this  $F_1$  generation of the hybrid. In this photograph (PLATE 4) the viable seeds may be readily identified by the dark color of their encasing glumes. The florets with white or straw-colored glumes did not develop seeds and later shriveled up and showed themselves completely sterile with no evidence of fertilization having taken place. In many cases, however, the feathery stigmas developed sufficiently so that they were extruded from the glumes, and in a less number of instances poorly developed stamens also showed outside the glumes. The cause of sterility appeared to reside more in a deficiency or the poor quality of the pollen than in an imperfect pistil, since many of the stigmas appeared very nearly normal in their development. Unfortunately a microscopic examination of the stamens and stigmas was not made, because of the pressure of other duties at that time.

The fertile flowers developed normally, becoming gradually more turgid as the seed filled out. Seven seeds were obtained from the panicle, five of



which were good-sized and plump, although smaller than the seeds of Dakota Amber sorgo. Two of the seeds were smaller and flatter than the others. The five seeds proved viable, and when planted produced vigorous  $F_2$  plants. A count of the florets on the panicle showed a possible 195 seeds, only 3.6 per cent of which produced seeds under greenhouse conditions, indicating in this case 96.4 per cent of sterility, but in other cases it has been very much less.

The  $F_1$  arose from a cross-pollinated seed taken from a sorgo plant, yet certain of the  $F_2$  plants grown in the open field at Chillicothe, Texas had rhizomes like those of Johnson grass and open, somewhat lax panicles, establishing beyond doubt the authenticity of the original cross. Other  $F_2$  plants showed differences, some being heavier-stemmed, like the sorgo parent, and practically devoid of rhizomes. Nearly all the plants had pithy stems, like Johnson grass, rather than the sweet juicy stem of the sorgo.

#### EXPLANATION OF PLATE 4

Panicle of an  $F_1$  plant of the cross between Dakota Amber Sorgo and Johnson Grass. Only the dark-colored (black) florets contain seed.





VINALI: PARTIAL STERILITY IN HYBRIDS





# STERILITY IN FRUITS: A SUMMARY OF TWENTY YEARS OF STUDY AT THE ROYAL HORTICULTURAL SOCIETY'S GARDENS

FRED J. CHITTENDEN

*Royal Horticultural Society's Gardens*

The experiments to which I am about to refer were begun in 1902 at Chelmsford, Essex, and have been continued since 1907 at the Gardens of the Royal Horticultural Society of England at Wisley, Surrey, first by myself and latterly by my colleague, Mr. Alan Rawes. They were started with the object of ascertaining what apples and pears, if any, could be safely planted in blocks of one variety, for it had been shown experimentally in England eighty years earlier that cross-pollination was necessary in certain pears, while, just before, Waite's work had called attention to the same need in American fruits. Later our work was extended to the other fruits hardy in the British Isles, except cherries with which we have made no experiments, and the intention of the experiments was extended to discover whether cross-pollination was more effective between one pair of varieties of pear or plum or apple than between another pair of varieties, and further, when the facts of partial or complete self-sterility had been established to discover the cause.

It seems best to confine my remarks to our work with apples and pears. Mr. Crane is dealing with somewhat similar experiments made at the John Innes Horticultural Institution, partly with apples but largely with plums and cherries, and it will therefore suffice to say that so far as plums are concerned, our results confirm the facts of self- and cross-sterility as found by him.

With English apples we have to deal with a great number of varieties of one group, viz., the large-fruited forms with little tannic content of the species *Pyrus Malus*, but it may be said that the comparatively small number of experiments we have made with forms of *P. Malus* outside the range thus defined, embracing varieties of "crab" apples both wild and cultivated seem to show that what is generally true of the one group of the species is true also of others. Our pears too belong to the single species *Pyrus communis*. Hybrids, in the sense of the term commonly accepted, do not enter into and complicate the question as they do in so many plants, and indeed into pears at least, in some parts of the world. Care has been taken in all our work to use authentic examples of the varieties with which we purported to be working. With some of our fruits, cherries and currants especially, nomenclature is often complicated by much synonymy, and identity is often obscure. Unless care be taken to identify correctly the material under experiment, much valuable time is lost and published results may be fallacious and

misleading. Apparently contradictory results not infrequently resolve themselves in the light of the probability of erroneous nomenclature.

Care has also been taken to make our experiments with plants in normal health, for abnormal conditions may either increase or diminish fruitfulness and thus give misleading results. At first we used trees growing in the open in our plantations for the pollination experiments, covering the flowers to be experimented upon with paper bags. The conditions inside a paper bag placed over a cluster of fruit, can scarcely be regarded as normal. So far as I know, no one has related the difference in temperature, air supply, and humidity inside such a bag in comparison with outside, but there must clearly be a considerable difference. Apart from this we found that while the protection afforded usually kept away all insect life and prevented chance pollination by wind, it was not always effective in shielding the flowers from adverse weather conditions, and many experiments were upset by this cause. Further the whole of the experiments had to be concentrated into a few days and we therefore carried out much of our later work upon both apples and pears (as well as plums) growing in pots and brought into a specially constructed glass house to flower. All insect interference is prevented by frames of wire gauze that cover the ventilators, and by disinfection and fumigation of the trees and house. Ants we find we cannot entirely exclude but these are prevented from reaching the blossoms by grease bands round the stems of the trees. We are thus able to extend the flowering period, make many more crosses since time is saved by the elimination of bagging, and avoid trouble from inclement weather.

In spite of these precautions against abnormality and against adverse weather during flowering time, it sometimes happens that the amount of fruit set is very small, even when the amount of flower is large. Thus, in one year, little more than 2 per cent of the crosses made on the apple Cox's Orange Pippin resulted in the setting of fruit, and it might have been concluded that this apple was infertile to the pollen of ten other well-known varieties, and fertile only to three. The average set, however, in other seasons has been over 34 per cent and this apple has set with the pollen of every variety so far tried. Our negative evidence was of no direct value.

When pollination is left to natural agencies the set of fruit is much below this even in the best years, and under the best conditions. When hand pollination is carried out under the best conditions and when what may be regarded as the most suitable variety is used for crossing the set of fruit rarely exceeds one-third of the flowers pollinated, and only very exceptionally reaches one-half. We thus have to reckon with failures following pollination experiments with these plants due to causes outside our control and more or less unknown.

While with the precautions we are able to take to prevent unwanted pollen from reaching the stigmas of the plants under experiment, we can feel confident that positive results have a definite meaning for us so far as the relation of pollen to ovule goes, when our crossings fail we can have no

such feeling of assurance. We can feel no confidence that a single negative result has any real meaning, nor even that repeated negative results may not be subsequently contradicted. Our long series of experiments has led us to conclude that negative results are in a large measure to be distrusted, but at the same time their constant repetition, and especially their occurrence alongside positive results when comparable crossings are made, do at least establish a probability.

The doubtful value of negative results comes in not only when we have to decide a question of self-sterility in these fruits but also when any attempt is made to compare the fertility of a variety to different pollens. Wide variations in the amount of fruit set may have no significance and only after similar results have been obtained in different seasons and on different trees, can negative results be taken as of real value.

If we seek an explanation of the prevalence of these negative results in nature as well as under experiment it may possibly be found in part in the developmental history of the flowers of apples and pears. We have made little investigation of pear flowers but something has been done by various workers in our laboratory with apples. We find that in England very frequently a small proportion of the pollen grains are obviously small, malformed, or abnormal in shape, and usually incapable of growth. This proportion in any flower does not usually exceed 10 per cent and may be less. On occasion a certain number of anthers may be completely aborted, but this is unusual. We have never seen in any variety in a normal year a smaller proportion than 70 per cent of apparently good pollen, and this relatively low figure was given by Ribston Pippin. Flowers of this variety from many localities gave a low reading for viable pollen as compared with other varieties. Whether the abortion of a portion of the pollen is a varietal character I am not in a position to say, but even in this extreme case it is certain that sufficient pollen is produced to furnish all the stigmas the plant can bear. I am inclined to think that the abortion of a certain amount of pollen and of many ovules may arise in another way.

In England the primordia of the flowers are laid down in summer, probably in some buds by the end of June, in others later, and a certain amount of development goes on up to October when it normally ceases, to be taken up again in January and continued until flowering time. Weather does not greatly affect the buds during the rest period, but it seems likely that when growth has again become active low temperatures may affect developments adversely. In January and February much of the cell division which finally results in the production of pollen grains and egg cells is in progress and goes on intermittently, being in all probability interrupted from time to time by sharp frosts, and it seems probable that at these times damage to developing pollen and egg cells is done, so that they cannot eventually carry out their proper functions. If this be so, and while our evidence is not absolutely conclusive the evidence we have leads us to believe it probable, then it is likely that the ovules of any one flower will suffer proportionately more than the



pollen. The anthers are of various ages and stages of development in a flower, while the (usually) ten ovules are of approximately the same age and at the same stage of development. Thus while the critical period of the pollen in any flower will be comparatively long that of the egg cells will be concentrated into a very short time. The occurrence of such bad spells of weather in England is intermittent and many ovaries may be affected while much pollen may escape.

In no variety of apple ordinarily cultivated have we found any approach to unisexuality in the flower. Such varieties occur but are not ordinarily cultivated. All varieties so far as we have seen produce under English conditions ample supplies of pollen equally in all flowers, and in no case have we found stigmas or ovary consistently aborted.

Our experiments have been made with nearly 150 varieties of apple, but confining ourselves to the 46 which have been used in the experimental house (and these include almost all varieties commercially grown) only one variety, Graham's Royal Jubilee, has failed to set fruit with its own pollen. In plantations, however, where it is one of the latest apples to come into flower, and where its flowering time scarcely overlaps that of any other commercially grown variety, it sometimes fruits very freely. I therefore conclude that this also is self-fertile. Mr. Crane has had, I think, one seedless fruit produced after self-pollination.

Apples may therefore be regarded as usually self-fertile, but both the kind and the degree of self-fertility differ.

*Self-fertility may be of two kinds.* In varieties such as Duchess of Oldenburg, White Transparent, and Golden Spire the fruits produced after self-pollination are, as a rule, seedless; though sometimes the testas are enlarged almost to their full size. This seedless type of fruit is, of course, normal for summer flowers of apples, where very rarely indeed is cross-pollination possible. It appears therefore that pollination itself has a stimulating effect upon growth whether followed by fertilization or not. Frequently after pollination enlargement of the embryo fruit begins, and may go on for some time, although the fruit never reaches its full development. In other cases, for instance in Beauty of Bath, self-pollination frequently results in seed production.

*Self-fertility also varies in degree.* Cox's Orange Pippin, for instance, has set only 4 fruits with us after self-pollination out of hundreds of self-pollinated flowers. It has set now and then with other workers, but with extreme rarity, and we have no evidence to lead us to suppose that there is more than one strain of this variety, one self-fertile, the other self-sterile. In the terms of the Mendelian Cox's Orange Pippin does not seem to be a mosaic: some other explanation of its occasional self-fertility must be sought. We may regard it as one of our least self-fertile varieties.

At the other end of the scale is the apple Rev. W. Wilks, which may be regarded as one of our most self-fertile varieties, and an instructive one. In various trials over a series of years an average of 18 per cent of



self-fertile fruits has been produced, the percentage in different years varying from 11 per cent to 41 per cent. When pollinated by other varieties (10) the average set has been 24 per cent, varying between 5 per cent and 71 per cent.

This difference of 6 per cent is of course not very great, and taken by itself it is perhaps not outside the bounds of experimental error, but taken in conjunction with figures obtained as the result of similar experiments with other marked self-fertile varieties, it is significant. In all such varieties we find that cross-pollination results in the production of greater crops than self-pollination.

It seems clear that not all the failures of pollination to secure fruit result from deficiencies in the ovules and it is very difficult to form a mental picture of the mechanism of self-sterility or such self-sterility as occurs.

We have some evidence that the effectiveness of pollen depends upon the rate of growth of the pollen tube. The pollen tube may not elongate sufficiently during the life of the style for fertilization to be effected. This may happen with any variety if the weather be so hot and dry as to shorten the life of the style in a marked manner, and it seems likely that the growth of the pollen tube is slower in its own style than in that of other varieties. If that be, the chance of foreign pollen to effect fertilization should be greater than that of own pollen. (In all references here "own pollen" means pollen of the same flower, or pollen of other flowers of the same tree, or pollen from flowers of other trees of the same variety.)

If this be so it is difficult to understand why. Feeding with sugar on the stigma does not improve the prospects of self-fertility so far as our experiments show, and it seems necessary to postulate the presence of some substance exerting a more or less inhibitory effect, and so far as we can see this inhibition is confined to the pollen tube growth of the same variety only for we cannot yet at any rate convince ourselves that, while there is certainly preference for the pollen of another variety, there is any preference for any other one variety above another. It is true that the percentage of fruit set by, say, Cox's Orange Pippin is greater with one variety of pollen than with another, so far as our experiments go, but there is so little consistency in the results obtained in different years that we cannot say there is any real evidence of cross-incompatibility between varieties of apples and of pears, as there certainly is between varieties of plums and especially of cherries. We must probably look to a study of this cross-incompatibility in cherries and plums to throw light upon the deeper problems of self-sterility in rosaceous fruits.

From time to time practical men suggest that this variety or that is the better pollenizer. For instance, Worcester Pearmain is said to be a good variety to plant with Cox's Orange and it proves experimentally to be effective, but the same has been claimed for the crab John Downie, but this has not behaved well under experiment. Our experiments have led us to conclude that it will be perfectly safe and equally effective to plant in proximity for

intercrossing any two varieties of European apples and pears the flowering times of which overlap.

In England we scarcely know the "burst of spring" which seems so characteristic of the American climate. Varieties do not follow in flower so quickly upon the heels of one another that all are past within a few days. Usually the flowering period of our apples is spread over several weeks so that the first is past long before the last begins. Choice of varieties for planting together is therefore restricted to those whose flowering periods most nearly coincide. Fortunately flowering period is a fairly fixed varietal characteristic, and the order of flowering scarcely varies from year to year, and that is true over the whole range of a variety's cultivation both at home and abroad. It has therefore been fairly easy to draw up reliable tables for the guidance of planters.

This is not the place to enlarge upon this aspect of the question nor upon the various side issues that have arisen around the problems since it was first attacked. After all our work we are still unsure of many of the factors that interfere with fruitfulness, but we have some guidance as to planting and we can with a certain amount of assurance fit any new variety into a planting scheme with faith that it will have an opportunity, if the scheme be followed, of giving its best results.

The general facts so far as we have given them for apples appear also to apply to pears.

Our experiments have been made with about a hundred varieties of pears and rather more than half of them have proved self-fertile in varying degrees. Many varieties have not yet set fruits with their own pollen, and while some of them at least will undoubtedly be found in the future to be self-fertile, some may be self-sterile, and most may probably be regarded as self-fertile in a minor degree. I speak thus guardedly in view of the suspicion with which, as I have pointed out, negative evidence must be regarded.

The same reduction of fertility as is found in apples on self-fertilization seems to prevail also in pears. For instance Conference has given 6 per cent of fruits from self-pollinated flowers, while flowers crossed on the same trees gave 15 per cent; Durondeau gave 5 per cent when selfed, 10 per cent when crossed; Marie Louise when selfed gave 10 per cent, when crossed 23 per cent, and so on.

Opportunities for crossing are therefore imperative with European pears as with apples, and as with apples, while I cannot speak too positively upon the point since we have made only a few thousand crosses and further work may bring to light some cases, no clearly marked instances of cross-incompatibility have been met with.

It may be of interest to notice that the only case of xenia (if this may be included in that group of phenomena) which we have come upon in the course of these experiments with our hardy fruits has been met with in pears.

Several varieties of pear when selfed produce seedless fruits and while the skin, flesh, and flavor are not different between the fruits produced as a

result of self-pollination and those following cross-pollination and which contain seeds, the shape is markedly different.

The carpels of the selfed fruits do not swell or rather increase in width, and consequently the diameter of these seedless fruits, usually in the upper half, is greatly reduced and we have an almost cylindrical instead of a pyriform fruit. Conference, normally a long pyriform fruit, shows this variation in a very marked fashion. Durondeau as seen in commerce is a fruit of frequently asymmetric form and investigation will generally show that the lack of symmetry is often correlated with the presence of one or more seedless carpels. Seedless apples on the other hand are not apparently affected in a similar way and may be as large and as typically proportioned as those which contain seeds. Asymmetric fruits certainly occur in apples and the lack of symmetry may at times be related to infertility of one or more carpels but is commonly due to some other cause, e. g., insect injury, or damage to one or more of the vascular bundles in the early stages of growth, sometimes brought about by frost.





# POLLEN PRODUCTION AND INCOMPATIBILITIES IN APPLES AND PEARS

RUDOLF FLORIN  
*Swedish Museum of Natural History*  
(WITH PLATES 5-7)

## INTRODUCTION

The sterility problem in fruit production is by no means a new one that has arisen in later years. As early as in the nineties attention was drawn to this question by scientific horticulturists in the United States. The works brought out by them have been the first of a whole series of similar studies, now assuming a scope which was hardly conceivable at an earlier period. This fact is mainly due to the immense importance of the sterility problem in the commercial fruit-growing, of which growers have become increasingly convinced.

The earliest investigations in the United States, in particular those of Waite (1894), were referred to in the Journal of the Swedish Pomological Society (Sveriges Pomologiska Förenings Årsskrift), but at that point matters remained for some time, and until 1913 no particular interest in these questions can be traced in Swedish horticultural journals. This year, however, in the journal mentioned there were published by Dahl and Sonesson (1913) tables showing the flowering seasons of different fruit varieties, compiled from numerous observations at different latitudes in Sweden during the years 1910-1912. At a meeting of the Swedish Pomological Society in Stockholm in 1914, Sonesson gave an account of particularly Waite's and the English investigations that had so far been published.

So a further year or two went by, and it was not until 1918 that growers began to be more seriously interested in the sterility problem. During the next few years the Pomological Society was, as before, responsible for the conduct of such investigations as were not undertaken on private initiative, and their operations were assisted by a small annual grant from the State. In the year 1923 the so-called "Permanent Committee for Orchard Research" (Permanenta Kommittén för Fruktdlingsförsök) was founded with the function of conducting all pomological experiments and researches in our country. This work has since been carried on at two stations, one in the neighbourhood of Stockholm in the garden of the Royal Academy of Agriculture at Experimentalfältet and one in Scania at the College of Agriculture, Alnarp, near Lund (Scania).

In spite of the slightly more favorable conditions, from the economic point of view, under which the Swedish experiments have been carried out since 1923, nevertheless the funds available are still inadequate for the

purpose. This circumstance should be taken into some account when reviewing the results of the Swedish activities in this sphere.

## INVESTIGATIONS ON PEARS

### TESTS OF SELF-INCOMPATIBILITY IN PEARS

The experimental study of self-sterility and cross-incompatibility in pears was begun by Waite (1894). This report on "The Pollination of Pear Flowers" has become almost a classic and has been repeatedly quoted in later works. Further, in the United States, Fletcher (1900 and 1911) and Powell (1902), and then later especially Ballard (1916), Tufts (1919) and Tufts and Philp (1923) have devoted exhaustive studies to the pollination problem in the pear.

In England various observations have been published by Hooper (1921) and Middlebrooke (1912), in Holland by Mrs. van Oijen-Goethals (1912-13 and 1916), in Switzerland by Müller-Thurgau (1903, etc.) and Osterwalder (1910) and in Germany by Ewert (1906, 1922, etc.). In Australia a short treatise on the same subject has been published by Prescott (1911).

In Sweden, finally, observations with regard to self-sterility in pears have been made by Stålfelt (1919 and 1920), N. Johansson (1921 and 1923) and E. Johansson (1926), while in 1925 Mrs. E. H. Florin published a detailed account of the pollination tests so far carried out with particular reference to the results obtained in central Sweden.

I have compiled in TABLE 1 those Swedish self-incompatibility tests in

TABLE 1  
SWEDISH TESTS OF SELF-INCOMPATIBILITY IN PEARS

VARIETIES	NUMBER OF BLOSSOMS POLLI- NATED	NUMBER OF BLOSSOMS SET	NUMBER OF FRUITS MATURED	PERCENT- AGE OF BLOSSOMS HAVING PRODUCED MATURE FRUITS	REMARKS
Alexander Lucas.....	189	8	6	3.2	Partially self-fruitful
André Desportes.....	401	4	..	0.0(?)	Self-unfruitful
Bartlett.....	247	1	0	0.0	Self-unfruitful
Belle Lucrative.....	778	26	9	1.2	Partially self-fruitful
Bergamotte Lübeck.....	508	...	38	7.5	Self-fruitful
Beurré Hardy.....	97	0	0	0.0	Self-unfruitful
Clapp Favorite.....	181	1	0	0.0	Self-unfruitful
Colorée de Juillet.....	396	17	0	0.0	Self-unfruitful
Comtesse Clara Frijs.....	249	9	6	2.4	Partially self-fruitful
Diamond Pear of Gothen- burg.....	100	6	2	2.0	Partially self-fruitful
Doyenné du Comice.....	563	13	1	0.2	Partially self-fruitful
Esperine.....	324	37	..	..	Partially self-fruitful(?)
Fondante de Charneu.....	342	23	16	4.7	Partially self-fruitful
Graf Moltke.....	527	19	13	2.5	Partially self-fruitful
Grise-Bonne.....	992	130	17	1.7	Partially self-fruitful
Herzogin Elsa.....	358	31	8	2.2	Partially self-fruitful
Hodge.....	261	2	0	0.0	Self-unfruitful
Hofsta.....	477	33	10	2.1	Partially self-fruitful
Jaune Hâtive.....	654	13	10	0.2	Partially self-fruitful
Pitmaston.....	146	0	0	0.0	Self-unfruitful
Président Drouard.....	153	0	0	0.0	Self-unfruitful
Soldat Laboureur.....	239	0	0	0.0	Self-unfruitful

which at least about 100 flowers have been involved. The table specifies the number of blossoms pollinated, the number of blossoms set, the number of fruits that reached maturity and the percentage of blossoms that produced mature fruits.

In our experimental studies we have always used the bagging method and branches or parts of branches have been enclosed in translucent white tissue-paper bags. The trouble with the bagging method is mainly that the light, temperature, and atmospheric conditions inside the bags are probably to a certain extent abnormal, with the result that blossoms and leaves may possibly fall off. However, as a rule we have been successful with not only pears and apples but also cherries and plums. Only certain apple varieties, as for instance the Swedish variety Åkerö, have sometimes caused us anxiety in this respect. The flowers have always been pollinated by hand in order to make the results as reliable as possible.

In order to avoid the possibility of mistake I have listed in the table the various varieties as "self-fruitful," "partially self-fruitful" and "self-unfruitful" respectively, since no account has been taken in our experiments of the existence in the fruits of seeds capable of germination (cf. Kraus, 1915, and Stout, 1916). Self-unfruitfulness is probably identical with complete self-incompatibility or at least with a high degree thereof, as the presence of seeds invariably stimulates the development of the adjacent tissues. On the other hand, a variety can be self-fruitful or partially self-fruitful, as the case may be, without at the same time being self-fertile, i. e., without forming seeds capable of germination, since the development of the fruit under certain conditions is independent of the stimulation of seeds. In the category "partially self-fruitful" I have placed varieties giving a positive result after self-pollination but manifestly producing a higher percentage of mature fruit after cross-pollination. A variety is only completely "self-fruitful" provided fruit-setting cannot be increased by cross-pollination. More or less complete self-sterility, therefore, occurs in pears to a greater extent than would appear from my table, and true self-fertility is of rare occurrence. Even in such a case as Bergamotte Lübeck, in which self-pollination of 508 blossoms produced 38 fruits, self-fertility apparently occurs only on a small scale, since such fruits as do result from self-pollination contain a preponderance of empty or shrivelled seeds.

In my opinion the so-called "self-fertility" which horticulturists have thought themselves to have proved in no small number of pear and apple varieties has, in many cases, to be regarded as not being true self-fertility but rather parthenocarpy or pseudocarpy induced by pollination. This may be a further reason why the results of self-pollination experiments have been so different in different districts that some authors describe a given variety as self-sterile while others classify it as at any rate partially "self-fertile." Another reason is that true self-fertility itself seems to be of a somewhat fluctuating nature.

Thanks mainly to the investigations carried out by Müller-Thurgau



(1898 and 1903), Ewert (1906-1910) and Osterwalder (1910) we know that obligate parthenocarpy exists neither in pears nor in apples, but that fluctuating facultative parthenocarpy does occur in these fruits. Certain changes in nutritive conditions and in the distribution of nutrients within the plant seem capable of replacing fertilization and inducing fruit development. About these conditions, however, we know too little to be able by means of fairly simple cultural practices consciously to induce parthenocarpy on a scale necessary to obtain a full crop from block plantings of self-sterile varieties.

In the comparatively few cases in which true self-fertility may be assumed to exist in pears and apples, even this quality is to a fairly large extent of a slight and fluctuating nature, and indeed is nothing to rely upon for purposes of commercial fruit-growing. The system of mixed planting, i. e., interplanting with pollenizers in order to ensure cross-pollination should therefore in most instances be employed in the orchards.

From the point of view of Swedish fruit-growing the most important result of the above-mentioned tests of self-sterility is the discovery that the majority, if not all, of the pear varieties hitherto investigated lack the capacity under ordinary conditions of setting fruit to a satisfactory extent with their own pollen. Consequently we have to mix up our plantings with suitable pollenizers.

In pears there is apparently no connection between the different degrees of self-fruitfulness and the viability of pollen. Complete pollen abortion does not occur in any variety. Among the self-unfruitful varieties we find on the one hand such a variety as Pitmaston with an average pollen germination of 8.0 per cent ( $\pm 1.9$  per cent), and on the other hand Soldat Laboureur with 63.0 per cent ( $\pm 3.8$  per cent). Among the partially self-fruitful varieties the power of pollen germination varies from 2.1 per cent ( $\pm 1.2$  per cent) to 83.0 per cent.

However, in selecting suitable pollinizers for commercially important varieties, it seems to be a matter of importance in the first place to choose good pollen-producers for that purpose, and I therefore propose to give now an account of our studies of pollen in pears.

#### PEAR POLLEN GERMINATION STUDIES

When I first began to take interest in the pollination problem of fruit-trees, about 8 years ago, the tests that had so far been carried out in Sweden concerned almost exclusively the occurrence of self-incompatibility. People had contented themselves with establishing the fact that several varieties required pollen from other varieties in order to produce fruit on a satisfactory scale, and it was thought that practically any variety would serve as a pollinizer provided it flowered at the same time as the variety whose flowers were to be cross-fertilized.

The correctness of this view was naturally open to dispute and it was therefore resolved to devote a close study to the pollination problem. There were in particular two questions which we desired to have answered: (1) to what extent does poor pollen exist in fruit varieties, and (2) what part is



played by physiological cross-incompatibility in the sterility of pears and apples. It is the first of these two questions that we shall concern ourselves mostly with now.

Germination tests with pollen of pears have been carried out in Europe particularly by Manaresi (1912), Kobel (1925), E. Johansson (1926), Mrs. Florin (unpublished) and myself (1920, 1921). Besides this, some sparse details have been published by Ewert (1906), Osterwalder (1910), Adams (1916), Passecker (1925) and others. As early as 14 years ago Manaresi published an account of particularly exhaustive germination tests with pollen from various kinds of fruit-trees, and the results obtained by him are reproduced to a large extent in the tables of pollen viability which I have compiled here. I have included all the information given in the various European works so far published which have seemed to be reliable and which deal with pollen that has been taken from orchard trees, i. e., not from greenhouse specimens nor such as have been influenced experimentally. In some few cases I have excluded certain particulars from the tables when I have not been sure as to which variety they concerned.

As a culture medium we have in most cases used a 10 per cent cane sugar solution. Comparisons have been made with agar-sugar media, but without obtaining any better results with them. The method of culture utilized gave excellent germination, affording apparently a reliable test for the relative viability of pollen. Kobel (1925) does not consider that the power of germination is a direct measure of the fertilizing ability of pollen, since those tubes which are formed in cultures with poor pollen often remain short and are sometimes more or less deformed. The fact that a pollen grain is in general capable of germinating in sugar solution does not necessarily mean that it can produce a tube long enough to reach the ovule in a flower even in the case of cross-compatibility existing. In certain cases this view seems to be fully justified and the percentage figures of germination may thus indicate a somewhat higher capacity of functioning than the actual. But in the main I regard the power of germination in sugar solution to be a very satisfactory gauge of the relative viability of pear and apple pollen.

The viability of pollen is in the first place a varietal characteristic. When in 1920 I wrote my first paper on pollen germination in pears and apples, I laid great stress on this point. Ewert (1921) on the other hand put forward the view that the power of germination of pollen in one and the same variety varies in a high degree from year to year. To a certain extent, indeed, this is in accordance with actual fact. In the subsequent germination studies carried out by Mrs. Florin and myself we have certainly been able to verify the correctness of my original conception, that each variety is characterized by a certain degree of pollen abortion, but on the other hand several varieties have shown themselves to be subject to a rather considerable variation in this respect. The pear varieties hitherto investigated in Europe may suitably, from the point of view of pollen viability, be divided up in the following way:

In this report of these tests "(1)" means that only one germination test

has been made; "(2)" means that two such tests have been carried out; in all other cases the results of at least three germination tests are known to me.

POOR POLLEN PRODUCERS AMONG PEARS  
AVERAGE POLLEN GERMINATION NOT ABOVE 30%

Alexander Lucas	Jargonelle
Baronne Leroy (1)	Knollbirne (1)
Bergamotte d'automne (1)	Louise Bonne de Printemps (2)
Bergamotte Lübeck	Maréchal de Cour (1)
Beurré d'Amanlis	Marguerite Marillat
Beurré Diel	Martin-Sec
Beurré Goubault (1)	Marxenbirne (1)
Beurré Mantecat	Mouille Bouche (1)
Bärikerbirne (1)	Olivier de Serres
Charles Cognée	Ottenbach's Schellerbirne (1)
Colmar d'Arenberg	Passe Crassane
Comtesse de Paris (1)	Pitmaston
Constant Lesueur	Précoce de Trévaux (1)
Fin de siècle (2)	Re Umberto
Fullerö (2)	Souvenir du Congrès
Gelbmöstler (1)	Theilersbirne (1)
General Tottleben (2)	Vicar of Winkfield
Glout Morceau	Virgoleuse
Graf Moltke	Wennström (1)
Grise-Bonne	Windsor (2)

Total: 40 varieties

MEDIUM POLLEN PRODUCERS AMONG PEARS  
AVERAGE POLLEN GERMINATION FROM 30 TO 70%

André Desportes	Eva Baltet (1)
Baronne de Mello (1)	Flemish Beauty
Bartlett	Fondante Thirriot (1)
Belle de Bruxelles	Hodge
Belle Lucrative	Jaune Hâtive (2)
Bergamotte Espéren	Joséphine de Malines
Beurré d'Angleterre (2)	König Karl von Württemberg
Beurré Clairgeau	Leboun's Butterbirne
Beurré Giffard	Le Lectier
Beurré Hardy (2)	Liegel's Honigbirne (1)
Blumenbach's Butterbirne (1)	Louise Bonne de Jersey
Bonne d'Ezée	Nouveau Poiteau
Cecilia	Président Drouard
Clapp Favorite	Radelma
Colorée de Juillet (1)	Rothärtler (1)
Doyenné de Comice	Rörstrand
Doyenné de Juillet (1)	Sarrasin (2)
Dr. Jules Guyot	Soldat Laboureur
Duchesse d'Angoulême	Späte von Ninove (2)
Duchesse de Berry d'Été	Tongre
Duchesse Grousset (2)	Torsö
Elizabeth (1)	Urbaniste (1)
Edmond Robitaille	

Total: 45 varieties

GOOD POLLEN PRODUCERS AMONG PEARS  
AVERAGE POLLEN GERMINATION AT LEAST 70%

August Pear (Experimentalfältet)	Gansel Bergamot (1)
Beurré Sterckmans (1)	Herzogin Elsa
Beurré Superfin (1)	Hofsta
Comtesse Clara Frijs	Jaminette (2)
Conference (1)	Madame Treyve (1)
Diamond Pear of Gothenburg (1)	Napoleon (1)
Easter Beurré (1)	Red Pear of Grenna (1)
Esperine	Reinholzbirne (1)
Eyewood (1)	Souvenir de du Breuil Père (1)
Fondante de Charneu (2)	Succès de la Millieraye

Total: 20 varieties

The list includes in all 105 pear varieties. Of these we find no less than 41 varieties in the group "Poor Pollen Producers," the average germinative power of which varies from 1 to 30 per cent. In these varieties the power of germination remains comparatively constant from year to year, though in exceptional cases it may nevertheless happen that one year one particular culture proves to contain a percentage of grains capable of germination

which deviates somewhat from the mean. Thus, for instance, this year (1926) there was obtained in one case a percentage of germination in Alexander Lucas of 12 per cent, while the mean amounts to 2.1 ( $\pm 1.2$ ). In order clearly to bring out the variability in the germination figures I have recorded in the pollen tables the lowest percentage of germination found and also the highest percentage, as well as calculated the mean error ( $\mu$ ) of the arithmetical mean by using the formula:  $\mu = \pm \sqrt{\frac{[\lambda]^2}{n(n-1)}}$  where  $[\lambda]^2$  means the sum of the squares of the actual deviations from the mean.

As "Medium Pollen Producers" I have listed those varieties whose mean percentage of germination is from 30 per cent to 70 per cent. To this group belong 45 varieties, but several of these are characterized by a high degree of variability.

The third group comprises such varieties as possess a power of pollen germination representing on an average at least 70 per cent, amounting in the material available to 20 varieties, or 19 per cent of the total number of varieties examined. This group is characterized by a fairly high degree of constancy in the percentage of germination from one year to another.

Pollen germinations for the three classes discussed above are illustrated in PLATE 5.

#### POLLEN MORPHOLOGY AND DEVELOPMENT IN PEAR VARIETIES

Even a passing glance at pollen cultures of pear varieties with a low percentage of germination will lead one to observe the irregular appearance of the pollen grains in them, particularly in comparison with varieties characterized by a power of germination of 70 per cent or more. It is of course very easy to assume that the irregular appearance of the pollen grains is in some way connected with the low germinative power of the pollen.

An examination of the pollen in Alexander Lucas, which usually germinates up to a maximum extent of only 2 per cent readily reveals that many of the pollen grains lack the first essential to enable them to germinate. They lack granular material and are empty. But even amongst those that prove to be filled with cytoplasm there are some that cannot germinate either. They are lacking in another essential for germination: they have not formed any germ pores. The following pollen categories occur in Alexander Lucas:

- a. Plump grains with 1-4 germ pores and containing granular material.
- b. Plump grains without germ pores but containing granular material.
- c. Plump grains without germ pores and more or less lacking in granular contents.
- d. More or less shrivelled grains without either germ pores or granular contents.

Out of 527 pollen grains especially examined only 24, or 4.6 per cent, proved to belong to the first category, which is the only one that includes grains partly capable of germination. As belonging to the second category 280 grains were counted, or 55 per cent; to the third 125 grains, equivalent to 23.7 per cent, and finally, to the fourth 98 grains, or 14.7 per cent. Put-

ting the last three categories together we find that no less than 93.4 per cent of the pollen grains are of a morphological character, making germination *a priori* inconceivable.

But even in the category that contains pollen grains capable of germination, irregularities are met with. The normal number of germ pores is three, but such regular grains are rare. Usually there are found even in this class more or less defective grains, either with four germ pores, which is comparatively unusual, or with less than three germ pores. Germination experiments with the pollen of Alexander Lucas extended over a period of four years and comprising in all ten tests, prove with desirable clearness that not quite 50 per cent of the pollen grains provided with germ pores are capable of forming pollen tubes.

Marguérite Marillat is another pear variety characterized by a very low germinative power of its pollen. Out of 554 grains counted, 93.7 per cent proved to lack the above-mentioned essential conditions to enable them to germinate, i. e., granular material and germ pores, while only 6.3 per cent fulfilled these conditions. In this case, too, however, germination experiments proved that only a portion of these 6.3 per cent actually are capable of forming pollen tubes.

Now, if we examine a variety possessing a relatively high power of germination, as e. g., Beurré Clairgeau, we find exactly the contrary state of things. It is true that approximately the same categories of pollen grains occur in this variety as in, for instance, Alexander Lucas, but their frequency is not at all the same. Pollen grains with germ pores and granular material were found this year (1926) to the extent of 88.8 per cent in the sample examined, while the other classes represented only 11.2 per cent together. The germination experiments resulted in 87 per cent of the pollen grains forming pollen tubes, that is to say, a result corresponding very well to the percentage figure for morphologically normal grains arrived at in the examination in lactic acid. In the course of years, however, it has been proved that Beurré Clairgeau belongs to that kind of pear varieties which is characterized by relatively variable power of pollen germination and it is therefore probable that the percentage of normal grains likewise varies in the same degree.

To what then is to be attributed this variation in the morphological appearance of the pollen and its low capacity of germination in such a variety as Alexander Lucas? Is the variability due to irregularities in the meiotic divisions of the pollen mother cells, as is the case, according to Shoemaker (1926), in such apple varieties as Stayman Winesap, or is the low germinative power to be ascribed to degenerative processes becoming active after the heterotypical and homoeotypical divisions have been completed, as was shown by Dorsey (1919) in the case of the plum?

In order to be able to answer this question already on this occasion, some time ago I cut through a number of buds of Alexander Lucas which had been fixed in Zenker's fixing fluid in 1923. The material was inadequate,



and the time too short to enable me satisfactorily to clear up the details regarding the chromosome conditions during the tetrad division. Further, the somatic chromosome number proved to be as high as about 56, which naturally rendered the interpretation of the heterotypical division difficult. What, therefore, I should like to state here is only the main features of the tetrad division in the pollen mother cells, which, however, are a matter of great interest in view of the high degree of pollen sterility occurring in several varieties of pears. I hope later on to have an opportunity of making a more detailed investigation into the cytology of some pear varieties in connection with the publication of the final account of our germination experiments with pear pollen.

The early prophase seems to show the appearance that is typical of the reduction division. From the diakinesis and metaphase stages (PLATE 6), however, it is evident that not all chromosomes have united into gemini but that some remain univalent. The univalent chromosomes arrive more slowly into the equatorial plane during the metaphase and stay longer there during the anaphase than the bivalent chromosomes. Apparently during the anaphase the univalents often undergo a longitudinal split, after which the halves move slowly toward the poles. Sometimes they reach the gemini-halves before these latter are enclosed by a nuclear membrane, and the immediate result of the heterotypical division, the two interkinetic nuclei, thus appear externally normal. But it often happens that the longitudinally divided univalents are delayed, either only slightly, or else more seriously, when they either remain lying somewhere between the two poles and are altogether enclosed by a nuclear membrane or else (8 in PLATE 6) give rise to several micronuclei. Sometimes this longitudinal division apparently does not take place, viz., when some univalent chromosome happens to lie in the periphery of the nuclear spindle (9 in PLATE 6). Even under these circumstances a chromosome can give rise to a micronucleus. Such figures as are shown at 6 in PLATE 6 indicate further that an uneven distribution of chromosomes between the daughter nuclei can take place during the anaphase.

In the homoeotypical division irregularities are still more conspicuous. The metaphase can appear fairly normal if no micronuclei have been formed previously. Sometimes there are, as in normal cases, two nuclear spindles in metaphase, but in addition scattered clusters of chromosomes in the cytoplasm. In some pollen mother cells, during the heterotypical division, a micronucleus forms between the two poles and, in addition, a number of chromosomes are ejected into the cytoplasm. These chromosomes do not divide further but probably degenerate, while the micronucleus, like the two larger nuclei, undergoes a homoeotypical division. In one case four nuclear spindles were observed in the metaphase of the homoeotypical division, two large and two small ones (10 in PLATE 6). The two microspindles were of course produced by two micronuclei formed during the heterotypical division.

Thus many irregularities occur in the distribution of the chromosomes,

in the formation of spindles and in the organization of nuclei. The tetrad has a normal appearance if the divisions have proceeded fairly regularly and the formation of micronuclei has not taken place or the latter have degenerated. But even in these frequently-occurring regular tetrads it is sometimes possible to discover initial degeneration in one or two of the microspores. If more than four nuclei are produced during the division of the pollen mother cells, traces of this are naturally forthcoming in the completed "tetrads." There are formed "tetrads" with 5-8 pollen cells, the size of which varies considerably.

Polyspory is thus a fairly usual phenomenon in Alexander Lucas. Polycary, on the other hand, seems to occur less often. Possibly a microspore may sometimes possess one large and one or more smaller nuclei. In isolated cases "tetrads" are met with containing only two large pollen cells. They seem, at least partially, to have originated from so-called regression nuclei (cf. Rosenberg, 1926).

The degeneration of microspores can sometimes be observed even before liberation, but as in the plum (Dorsey, 1919) suppression takes place principally after the liberation of the microspores. Occasionally the greater part of an anther loculus degenerates already at an early stage.

From what has already been mentioned regarding the tetrad division of the pollen mother cells in Alexander Lucas this process evidently closely recalls the tetrad division in certain hybrids between gametes possessing unequal chromosome numbers. The quality of my material does not permit me as yet to make any detailed comparison with plants and in particular Rosaceae previously described as distinguished by irregular chromosome distribution during meiotic divisions (Rosenberg, 1917; Täckholm, 1922; Kihara, 1924, etc.). It is very probable, however, that the different varieties of pears are characterized by a very different cytological constitution.

We are certainly justified in drawing the conclusion that the primary cause of the low germinative power of pollen in 38 per cent of the pear varieties so far investigated in Europe is to be found in the irregular chromosome distribution during meiosis. This seems further to hold good in the case of at least some of the medium pollen producers as well.

The most remarkable feature of this latter group, however, is the variation in the germinative power of the pollen from year to year and even in different tests within the same blooming period in many cases being considerable. External factors probably cause this variability. Within the last years a number of investigations on the disturbing influence of external factors on the meiotic divisions have been carried out, a problem obviously being of the utmost theoretical and practical importance. I am here naturally disregarding the effects of narcotics, Röntgen and radium rays, chloral hydrate, etc., and am thinking exclusively of such factors as, apart from hybridization, may influence the meiotic divisions, outside the scope of scientific experiment. De Mol has recently shown that abnormal pollen with an augmented number of nuclei is produced by what he calls physiological stimuli.

It is, however, difficult to form an idea of the causes of the disturbances and determine whether De Mol's results have any direct interest in the case of those cultivated plants with which we are dealing here.

Of more immediate interest in this connection are the works which deal with the influence of cold on the meiotic division in the pollen mother cells.

Miss Borgenstam (now Mrs. E. H. Florin) (1922) has investigated the meiotic divisions in the pollen mother cells of *Syringa chinensis*. She examined cytologically one and the same specimen of this species, firstly without any special treatment and second after freezing the buds. When the buds had not undergone a process of freezing, the meiotic divisions appeared on the whole to proceed normally. In the diakinesis of the heterotypic division all the 40 chromosomes were fused into gemini and in the metaphase these arranged themselves regularly in the equatorial plane. The anaphase took a normal course, as did the interkinesis and the subsequent homoeotypic division. On the whole there appeared no delayed chromosomes or reconstruction of the nuclei.

Entirely different nuclear phenomena were obtained after twigs of one and the same specimen had been exposed to a temperature of 0° C. As early as in the "resting" condition the nuclei of the pollen mother cells showed several anomalies and cells showing various phenomena of degeneration were common. Nuclei being capable of continued development behaved in different ways in the diakinesis stage. Many contained twice as many gemini as under normal conditions, i. e., 40. This abnormality proved to have arisen in pollen mother cells having contained two more or less fused haploid nuclei. During the final division in the archesporial cells, which normally leads to the formation of pollen mother cells, the wall formation had thus been inhibited. In the metaphase condition giant spindles were obtained showing numerous scattered chromosomes, some of which failed to reach the poles during the anaphase before the nuclear membranes began to form. Sometimes the reduction division proved to have ceased in an early stage and a regression-nucleus had been formed, of whose further fate, however, we are entirely ignorant. The homoeotypic division, too, of course proceeded irregularly and delayed chromosomes appeared. The result of the division of the pollen mother cells was in these cases irregular tetrads, containing either several micronuclei or only two large microspores.

Michaelis (1926) recently investigated the influence of cold on the quality of pollen and its development in two species of *Epilobium*. In the case of *Epilobium angustifolium* he found in specimens which had not been influenced experimentally, but rarely pollen grains with four germ pores and the majority of the pollen grains possessed the normal number, viz, three germ pores. After a specimen of this kind had undergone freezing, the anthers subsequently matured proved to contain a considerably higher percentage of four-lobed pollen grains, and in addition, pollen grains with more than four and less than three germ pores. In the frozen pollen there further appeared a large number of shrivelled empty grains. In *Epilobium hirsutum* Michaelis



did not find under normal conditions a single pollen grain with four germ pores, but in the specimens exposed to freezing there appeared numerous disturbed tetrads.

The cytological investigation then carried out showed the nuclei of the pollen mother cells to be extremely sensitive during the period that elapses between the synapsis and the diakinesis stage. In diakinesis the gemini-formation proved to be very incomplete and possibly the pairing of the chromosomes may sometimes not take place at all. In the course of the subsequent development there appeared numerous irregularities, which in certain cases resulted in diploid regression nuclei. During the anaphase it often happened that several chromosomes remained outside the two daughter nuclei, the latter not infrequently proving to be of unequal size and containing a varying number of chromosomes. Through all these anomalies the tetrads naturally got a very irregular appearance, and Michaelis was able to show dyads as well as "tetrads" with more than four microspores in each.

Michaelis maintains that in general hybrids must be assumed to be more sensitive to disturbances through external influences than pure species. Those pear varieties which in the course of our germination experiments constantly show a low percentage of pollen germination probably constitute hybrids between parents possessing different chromosome numbers and are characterized by an exceptionally disturbed meiosis. An additional disturbance caused by the influence of external factors, as for instance cold, certainly does not pass them by without leaving some trace, but owing to the high pollen sterility already existing before on constitutional grounds, the result is less felt in the germination cultures. On the other hand, in varieties characterized by varying capability of pollen germination, the tetrad division probably proceeds under suitable conditions without any large disturbances, while unfavorable external factors can easily bring about a fairly important aggravation of sterility.

Finally, those varieties which are constantly characterized by a high degree of pollen viability probably consist generally of hybrids between nearly related parents possessing the same number of chromosomes. They seem preponderatingly to show regular meiosis and are therefore relatively insensitive to the influence of external factors.

#### TESTS OF CROSS-INCOMPATIBILITY IN PEARS

As in the case of the self-pollination tests, the Swedish cross-pollination experiments have been carried out on orchard trees by the use of the bagging method. In all cases where this was necessary the flowers were emasculated in order to obtain decisive results. Pollination was usually effected with the greatest care by hand in this way: the operator seized a stamen with a pair of pincers and then touched the surface of stigmas with the contents of the anther. This we consider a more reliable method than to use a brush.

Having carried out our investigations concerning unfruitfulness in fruit varieties, principally from the point of view of the mature, marketable fruit,



TABLE 2  
SWEDISH TESTS OF CROSS-INCOMPATIBILITY IN PEARS

SEED PARENT	X	POLLEN PARENT	NUMBER OF BLOSSOMS POLLINATED	NUMBER OF BLOSSOMS SET	NUMBER OF FRUITS MATURED	PER- CENTAGE OF BLOSSOMS HAVING PRODUCED MATURE FRUITS
Alexander Lucas	x	Clapp Favorite.....	158	24	21	13.3
Alexander Lucas	x	Flemish Beauty.....	109	18	16	14.7
Alexander Lucas	x	Président Drouard.....	146	29	28	19.2
Belle Lucrative	x	Comtesse Clara Frijs...	239	58	35	14.6
Belle Lucrative	x	Hofsta.....	251	55	42	16.7
Belle Lucrative	x	August Pear (Experi- mental-fältet).....	224	46	41	18.3
Colorée de Juillet	x	Belle Lucrative.....	200	26	10	5.0
Comtesse Clara Frijs	x	Diamond Pear of Gothen- burg.....	120	41	38	31.7
Comtesse Clara Frijs	x	Belle Lucrative.....	223	105	78	35.0
Comtesse Clara Frijs	x	Esperine.....	130	50	49	37.7
Diamond Pear of Gothenburg	x	Doyenné Boussoch.....	100	1	0	0.0
Diamond Pear of Gothenburg	x	Bergamotte Lübeck....	90	15	11	12.2
Diamond Pear of Gothenburg	x	Bartlett.....	145	50	24	16.5
Doyenné du Comice	x	Doyenné Boussoch.....	210	2	1	0.5
Doyenné du Comice	x	André Desportes.....	166	6	4	2.4
Doyenné du Comice	x	Esperine.....	190	19	7	3.7
Doyenné du Comice	x	Diamond Pear of Gothen- burg.....	180	8	8	4.4
Doyenné du Comice	x	Belle Lucrative.....	187	78	17	9.1
Esperine	x	Comtesse Clara Frijs...	230	23	8	3.5
Esperine	x	Belle Lucrative.....	268	45	24	8.9
Fondante de Charneu	x	André Desportes.....	214	32	24	11.2
Graf Moltke	x	Président Drouard.....	95	10	10	10.5
Graf Moltke	x	Belle Lucrative.....	247	95	83	33.6
Grise Bonne	x	Louise Bonne de Jersey	289	161	55	19.0
Grise Bonne	x	Rörstrand.....	534	137	116	21.1
Grise Bonne	x	Belle Lucrative.....	497	152	126	25.3
Grise Bonne	x	Esperine.....	275	184	122	44.4
Herzogin Elsa	x	Jaune Hâtive.....	320	74	41	12.8
Herzogin Elsa	x	Bartlett.....	271	95	38	14.0
Herzogin Elsa	x	Bergamotte Lübeck....	114	44	16	14.0
Hovsta	x	Herrö.....	313	138	78	24.9
Joséphine de Malines	x	Belle Lucrative.....	207	77	27	13.0

in the great majority of crosses we have used as pollen parents varieties producing pollen of good, or at least, medium quality. If then, under such circumstances we find a case in which a certain cross has given a negative result, this is obviously caused by physiological incompatibility between the two varieties in question.

It may at once be pointed out that we have not yet come across a single definite case of physiological inter-incompatibility in pears. It is true that in the above cross-pollination table two crosses have been noticed in which the percentage of blossoms that produced mature fruits is 0 and 0.5 respectively. In both cases Doyenné Boussoch is the pollen parent. Unfortunately I do not know the quality of the pollen in this variety either from Swedish tests or from tests made in other countries, and I am therefore unable to determine whether true cross-incompatibility exists in these cases or whether the nearly

complete cross-sterility is due to imperfect pollen. But in view of our experiences otherwise I think the latter explanation is the more likely one.

Further, the question may be asked whether different degrees of cross-compatibility occur which would render it necessary for practical purposes to recommend certain definite combinations of varieties. This question is not at all an easy one to answer on the comparatively meager material which has so far been brought together in Sweden. Even a cursory glance at TABLE 2 will suffice to draw attention to the varying percentages of blossoms that have produced mature fruits, which are of course chiefly to be ascribed to the different fruiting habit in different varieties. Disregarding some few exceptions, however, the percentage figures for each female parent often prove to be remarkably similar. Thus, the percentage of mature fruits in three crosses made with Alexander Lucas as female parent varies only between 13.3 and 19.2, in the case of three crosses with Belle Lucrative only between 14.6 and 18.3, in three crosses with Comtesse Clara Frijs only between 31.7 and 37.7, and in three crosses with Herzogin Elsa as female parent only between 12.8 and 14. If I am right as regards the power of pollen germination in Doyenné Boussoch our cross-pollination experiments on the Diamond Pear of Gothenburg do not represent any exception to this rule, since in the two other crosses 12.2 and 16.5 per cent of the blossoms respectively produced mature fruits. I am not yet able to express an opinion as to how the results of the crosses with Doyenné du Comice as female parent are to be interpreted. Excepting the cross Doyenné du Comice  $\times$  Belle Lucrative, these crosses have not been very successful, although two of the varieties used as male parents possess good pollen and one produces pollen of medium quality. The cross-pollinations with Graf Moltke as female parent seem to constitute an exception from the rule just indicated but even this one loses its indisputability when I state that the cross Graf Moltke  $\times$  President Drouard, which gave 10.5 per cent mature fruits, was carried out on an espalier-tree, while the cross Graf Moltke  $\times$  Belle Lucrative, which gave 33.6 per cent mature fruits, was effected on an ordinary standard tree. Finally, the crosses on Grise-Bonne likewise depart from the rule, but in this case too the conditions are complicated. The crosses with Rörstrand and Belle Lucrative as pollen parents were carried out in one and the same year and gave approximately the same result. The other two crosses were made in another year likewise on one and the same tree and under as uniform conditions as possible but gave quite different percentages of mature fruits, namely 19 and 44.4 respectively. Here, however, it has to be observed, that Louise Bonne de Jersey has a comparatively poor pollen—the mean percentage of germination found in this variety being 38.7 ( $\pm 4.7$ ), while the pollen in Esperine is viable to an average of 82.4 per cent ( $\pm 3.3$ ). Further, Louise Bonne de Jersey is very sensitive to low winter temperatures at such a high latitude as that of Stockholm, and as a matter of fact this variety had suffered severely during the winter preceding the flowering season in the course of

which the last-mentioned crosses were made. A pollen germination test carried out at the same time gave a result of only 22 per cent viable pollen.

Thus, in the Swedish cross-pollination tests no widely differing degrees of physiological cross-compatibility have so far been met with. However, account must be taken of the fact that the data so far collected are not very numerous. When studying the illuminating tables published by Tufts and Philp (1923) we find several cases of considerable differences in compatibility in crosses made with one and the same variety as female parent without it being possible to ascribe these differences to varying ability on the part of the pollen parents to produce viable pollen.

Judging from the American as well as the Swedish tests cross-incompatibility in pears does not exist, or is, at any rate, of rare occurrence in pears. It is also manifest that even in more or less self-fruitful varieties the fruit-crops can be considerably augmented by means of cross-pollination, particularly if varieties producing a considerable amount of viable pollen are used as pollen parents. In planning orchards intended to be commercially profitable these circumstances should be given close attention.

## INVESTIGATIONS ON APPLES

### TESTS OF SELF-INCOMPATIBILITY IN APPLES

The experimental study of self-incompatibility in apples was begun in the United States about 35 years ago. The earliest of the investigators were Waite (1898), Fletcher (1900), Powell (1902) and Close (1903). Since then a long series of American scientists have occupied themselves with the sterility problem in the apple, namely: Lewis and Vincent (1909), Alderman (1918), Wicks (1918), Overholser (1919), Gowen (1920), Morris (1921), Overholser and Jacob (1921), Sax (1921 and 1922), Auchter (1922), Crandall (1922), Dorsey (1922), Overholser and Cameron (1922), Auchter (1923), Keil (1923), Wellington (1923 and 1924), Auchter and Schrader (1926) and MacDaniels (1926).

From Canada I am aware of only two works dealing with self-sterility in apples, viz, one by Logsdail (1917) and one by Macoun (1923).

In Europe, Müller-Thurgau (1905 and 1908) in Switzerland was the first to interest himself in the fertilization of the flowers in orchards. At about the same time Ewert (1906) in Germany brought out his work on the biology of the flowers of different fruit-trees. In the same country, again, Zacharias (1911) has devoted a certain amount of attention to the carrying out of self-sterility tests with apples, particularly Gravenstein. In Holland and Denmark a few small investigations have been made by Mrs. von Oijen-Goethals (1912-1913) and Lindhard (1919) respectively.

In England horticulturists have devoted much attention to the question of the occurrence of self-sterility in apple varieties and accounts of experiments have been published by Backhouse (1912), Chittenden (1914), Middlebrooke (1915), Miss Sutton (1918), Hooper (1921, etc.), Rawes (1922) and Crane (1923).

TABLE 3  
SWEDISH TESTS OF SELF-INCOMPATIBILITY IN APPLES

VARIETIES	NUMBER OF BLOSSOMS POLLI- NATED	NUMBER OF BLOSSOMS SET	NUMBER OF FRUITS MATURED	PERCENT- AGE OF BLOSSOMS HAVING PRODUCED MATURE FRUITS	REMARKS
Arreskov.....	462	6	2(?)	0.4(?)	Partially self-fruitful(?)
Beauty of Bath.....	268	8	3	1.1	Partially self-fruitful
Belle de Boscoop.....	702	22	18	2.6	Partially self-fruitful
Bismarck.....	913	4	4	0.4	Partially self-fruitful
Boiken.....	187	12	..	..	Partially self-fruitful(?)
Brunnsäpple of Halland...	215	38	8	3.7	Partially self-fruitful
Cox Orange.....	344	1	1	0.3	Partially self-fruitful
Cox Pomona.....	558	3	..	0.0(?)	Self-unfruitful(?)
Duchess of Oldenburg....	513	32	..	0.0	Self-unfruitful
Ecklinville Seedling.....	260	0	0	0.0	Self-unfruitful
Fiery Red Pigeon.....	214	58	11	5.1	Partially self-fruitful
Gelber Richard.....	149	1	..	0.0(?)	Self-unfruitful(?)
Gladstone.....	408	0	0	0.0	Self-unfruitful
Golden Noble.....	145	0	0	0.0	Self-unfruitful
Graham's Royal Jubilee...	190	0	0	0.0	Self-unfruitful
Gravenstein.....	261	10	..	..	Partially self-fruitful(?)
Gruschevka Krasnaja....	607	7	1	0.2	Partially self-fruitful
Hawthornden.....	262	8	..	..	Partially self-fruitful(?)
James Grieve.....	554	5	0	0.0	Self-unfruitful
King of the Pippins.....	106	1	..	0.0(?)	Self-unfruitful(?)
Kutofsky Nalivia.....	312	27	..	..	Partially self-fruitful(?)
Maglemer.....	360	0	0	0.0	Self-unfruitful
Mank's Codlin.....	248	9	..	..	Partially self-fruitful(?)
Mère de Ménage.....	371	10	5	1.3	Partially self-fruitful
Oranie.....	411	0	0	0.0	Self-unfruitful
Princess Noble.....	272	0	0	0.0	Self-unfruitful
Prinz.....	161	2	0	0.0	Self-unfruitful
Ringstad.....	308	3	1	0.3	Partially self-fruitful
Ribston.....	583	9	7	1.2	Partially self-fruitful
Rother Eiserapfel.....	169	0	0	0.0	Self-unfruitful
Signe Tillisch.....	114	0	0	0.0	Self-unfruitful
Steklianka Kremera.....	610	38	1	0.2	Partially self-fruitful
Stenkyrke.....	368	1	0	0.0	Self-unfruitful
Svanetorp.....	162	2	0	0.0	Self-unfruitful
Säfstaholm.....	807	0	0	0.0	Self-unfruitful
William's Favorite.....	211	0	0	0.0	Self-unfruitful
Yellow Newtown.....	419	0	0	0.0	Self-unfruitful
Yellow Transparent.....	607	25	7	1.2	Partially self-fruitful
Åkerö.....	153	0	0	0.0	Self-unfruitful

Finally, in Sweden tests of self-incompatibility in apples have been undertaken by Sonesson (1914), Stålfelt (1919 and 1920), Lindfors (1922), N. Johansson (1921 and 1923), Mrs. E. H. Florin, E. Johansson (1926) and myself (1918).

As in the case of the statistics for the pears, TABLE 3 indicates the number of blossoms pollinated, the number of blossoms set, the number of fruits matured and the percentage of blossoms that produced mature fruit, and contains data regarding 39 varieties. The results of the Swedish investigations agree quite well with those obtained in other countries, since more than half of the varieties examined seem to be completely self-sterile and the rest in



most cases only very slightly self-fruitful. Just as with the pears, the degree of self-fruitfulness is entirely independent of the quality of the pollen, and even several varieties with poor pollen have proved to be partially self-fruitful in our pollination tests.

It appears, however, that this self-fruitfulness is probably only in few cases to be regarded as true self-fertility, since many apple varieties show a tendency to parthenocarpy (Müller-Thurgau, 1908; Ewert, 1909; etc.). This tendency, however, cannot for the present be utilized from the commercial point of view, since in most varieties parthenocarpy is of a weak and fluctuating nature and the physiological conditions causing parthenocarpic development are far too little understood. Thus, all apple varieties hitherto examined in Sweden require cross-pollination and cross-fertilization in order to give satisfactory crops.

Before proceeding to give an account of our tests of cross-incompatibility in apples, I propose to touch on the question of pollen viability in different apple varieties.

#### APPLE POLLEN GERMINATION STUDIES

As in the case of pear pollen, Manaresi (1912) was the first researcher in Europe to publish a detailed account of germination studies with regard to apple pollen. Some scattered observations had, however, already been made by Ewert (1906) and Osterwalder (1910). Further, since 1912 Adams (1916), Kobel (1925) and Passecker (1926) have in Central and Western Europe contributed to our knowledge of pollen viability in different varieties. As regards America, there have been investigations made by Knight (1917), Martin and Yocum (1918), Auchter (1921), Beaumont and Knight (1922), MacDaniels (1925), Knowlton and Sevy (1925), etc. In Sweden this study has been carried on on a fairly large scale, having been begun in 1918. The first detailed account was published by me in 1920 and a further contribution in 1921. Since then, besides myself, Mrs. E. H. Florin (1925) and E. Johansson (1926) have been engaged in pollen germination studies. The table which I have compiled for this occasion represents a summary of all investigations published or carried out in Europe of which I have a close knowledge.

As in the case of pear pollen we have used a 10 per cent cane sugar solution as a culture medium, and have obtained very good results. (See PLATE 7.)

From the point of view of pollen germinability it would seem that the apple varieties so far investigated might suitably be divided into three groups, as in the following list. In this list and in all following lists (1) means that only one germination test has been carried out; (2) means that two such tests have been made; in all other cases the results are from at least three germination tests.

## POOR POLLEN PRODUCERS AMONG APPLES

AVERAGE POLLEN GERMINATION NOT ABOVE 30%

Allington	Red Gravenstein
Baldwin	Reinette Canada
Belle de Boscoop	Reinette Harbert
Blenheim	Rheinischer Bohnapfel (2)
Bossanka	Rhode Island Greening
Bramley's Seedling (2)	Ribston
Calville des Femmes (1)	Riesenboikenapfel (2)
Doctor Nansen	Roter Eiserapfel
Flintinge	Rotkantzler (1)
Frösåker	Souvenir de l'Éveque (2)
Gascoyne's Scarlet (2)	Spelling Favorite (1)
Gravenstein	Stäfner Rosenapfel (1)
Grüner Fürstenapfel (1)	Warner's King
Hambling's Seedling	Weisser, flammiger Kardinal (1)
Langeland (1)	White Spanish Reinette
Martin Becker (2)	Winterzitronenapfel (1)
Mecklenburger Königsapfel (1)	Yellow Newtown
Mère de Ménage	

Total: 35 varieties (= 14%)

## MEDIUM POLLEN PRODUCERS AMONG APPLES

AVERAGE POLLEN GERMINATION FROM 30 TO 70%

Anna Stina (1)	Peasgood's Nonsuch (2)
Aspa (1)	Perlenreinette (1)
Beefing, striped (1)	Pfirsichroter Sommerapfel (2)
Björkvik (1)	Pitmaston Pineapple (1)
Calville rouge d'hiver	Postophe d'été (1)
Champaigne Reinette	Red Prinz (1)
Court-pendu-plat	Reinette grise (1)
Drap d'Or (1)	Reinette Wormsley (1)
Gotthard (2)	Ringstad
Himbeerapfel von Holowaus	Roter Ananasapfel
Hörningsholm's Rosenapfel	Schöner von Nordhausen (1)
Jensen's Kaniker (1)	Simkulja (1)
Julity Kutofsky	Springgrove Codlin (2)
Klarapfel (Eneroth)	Stenkyrke
Kutofsky nalivia	Södermanland (2)
Mannington's Pearmain	Titovka
Mecklenburger Kantapfel (1)	Tobiasler (1)
Mölleskov (2)	Transparente de Croncels
Northern Spy (1)	Wealthy
Ontario	White Astrachan
Palasatœ nalifnoe (1)	Winter Banana
Parker's Pippin (1)	Värnanäs

Total: 44 varieties (= 17%)

## GOOD POLLEN PRODUCERS AMONG APPLES

AVERAGE POLLEN GERMINATION AT LEAST 70%

Adams Pearmain (1)	Krusenberg (2)
Adersleber Kalvill. (1)	Krüger's Goldreinette (1)
Alexander	Lady Sudeley (1)
Alfriston (2)	Lambron (2)
Alnarp's Winterstreifling (1)	Langer, roter Himbeerapfel (1)
Alnô (1)	Langley Pippin (2)
Antonovka	Langton's Nonsuch
Apfel von ülsen	London Pippin
Arreskov (2)	Lord Derby (1)
Arvid	Lord Grosvenor (2)
Astrakaner, grosser, klarer	Lord Suffield
Bananas (Eneroth) (1)	Lothringer (1)
Beauty of Bath	Maglmer
Beechamville's Seedling (1)	Mank's Codlin (1)
Berner Rosenapfel	Margaretta
Betty Geeson	Margil
Bismarck	Menigasker (1)
Bodil Neergaard (2)	Minister von Hammerstein
Boiken	Minnesota (1)
Borovinka	Mr. Leopold de Rothschild
Borsdorfer, gestreifter, böhmischer (1)	Odenwälder (2)
Broholm's Rosenapfel (1)	Okabena (2)
Brunnsäpple of Halland	Oranie
Calville blanche d'hiver	Paeregaard (1)
Canada Baldwin (1)	Papirovka (2)
Carpentin (1)	Pearmain Schwarzenbach (1)
Cellini	Pennington (2)
Charles Ross (2)	Pewaukee (1)
Chelmsford Wonder	Pigeon St. Louis (1)
Chenango (1)	Pigeon Wöldike (1)
Cludius' Herbstapfel (1)	P. J. Bergius
Coe's Golden Drop (1)	Plodovitka (2)

Collins	Prince Albert (1)
Cox Orange	Princess Noble (2)
Cox Pomona	Quetier
Danziger Kantapfel	Red Astrachan
Degeneapfel (2)	Red Margareta (1)
Domine (1)	Reinette d'anas
Dronning Louise	Reinette Baumann
Dutchess of Oldenburg	Reinette Berk
Duke of Devonshire (1)	Reinette de Granville (1)
Dumelow (1)	Reinette Oberdieck (1)
Early Rivers (1)	Reinette d'Orléans
Early Victoria (2)	Reinette Pederstrup (1)
Ecklinville Seedling (1)	Reinette Zuccalmaglio (1)
Edelborsdorfer (1)	Risäter (1)
Edelgraeuch (1)	Rival (2)
Edelrambour von Winnitza (2)	Rosenhäger (2)
Geh. Dr. Oldenburg	Rosmarin (1)
Edward VII (1)	Roter Jungfernapfel
Elise Rathke (1)	Rudolph's Liebling (1)
English Golden Pippin (1)	Rudolph's Zwiebel-Borsdorfer (1)
Fall Wine	St. Lawrence (2)
Fameuse (2)	Salatorewka (1)
Fiery Red Pigeon (2)	Sauergraeuch (1)
Fiesser's Erstling	Scarlet Pearmain
Filippa	Schwedischer Winterpostoph
Fleiner du roi (2)	Signe Tillisch
Flädie	Skovfoged (2)
Folkestone	Skvosnoj nalif (1)
Frass' Sommerkalvill	Sparreholm
Freiherr von Berlepsch (2)	Sperher (1)
Gelber Richard	Steklianka Kremera (1)
Gladstone (2)	Stensberg (1)
Glanzeinette (2)	Sturmer Pippin (1)
Golden Noble	Stäringe
Graham's Royal Jubilee	Suislepper (1)
Grosse Casseler Reinette (1)	Svanetorp (1)
Grossherzog Friedrich von Baden (2)	Sweet Bough (1)
Grushevka Krasnaja	Sykehouse (1)
Guldborg (1)	Säfstaholm
Hanaskog (2)	Tettowo (2)
Hawthornden	The Senator
Hedenlunda	Thomas Rivers (2)
Hohenstaufen's Rosenapfel (1)	Thurgauer Weinapfel (1)
Hurlbut (1)	Tjernoguse czerwone (1)
Irish Peach (1)	Tosterup (1)
James Grieve (2)	Tranekjaer (1)
Jonathan	Ullerud (1)
Jämshög	Waldhöfler (1)*
Kaniker (Scanish)	Vaterapfel ohne Kern (1)
Karabowka (2)	Villand (1)
Kavlås (2)	Vineuse Rouge
Kerry Pippin (1)	William's Favorite (2)
Keswick Codlin	Worcester Pearmain (1)
King of the Pippins	Yellow Bellflower
Kleiner Langstiel	Yellow Transparente
Kronprinz Rudolph von Österreich (1)	Akerö

Total: 176 varieties (= 69%)

The list comprises in all 255 apple varieties. Out of these 35 varieties, or 14 per cent, are included in the group "Poor Pollen Producers," in which the mean percentage of pollen germination is at the highest 30 per cent. In this group the power of germination is in most cases comparatively constant from year to year, although in isolated cases a sample may deviate rather considerably from the average. Thus, for instance, in one sample of Gravenstein there were obtained, in 1918, 17 per cent of viable pollen, while the mean of germination power for this variety is 7.3 per cent ( $\pm 1.3$ ). With Gravenstein we have carried out a large number of pollen germination tests in Sweden. Our experiences regarding the unsuitability of this variety as a pollen parent seem to be shared both by Central European and by American authorities.

As "Medium Pollen Producers" I have noted those varieties whose mean percentage of germination ranges between 30 and 70. To this group belong 44, or 17 per cent of the varieties so far examined. Many of them

are characterized by a high degree of variability in the power of pollen germination.

The third group comprises varieties possessing a germinative power of, on an average, at least 70 per cent, in the material available representing no less than 176 varieties, or 69 per cent of those investigated. Pollen viability in many of these varieties is remarkably constant.

A comparison between the results of the pollen germination studies with pear and apple pollen shows that more or less poor pollen production is relatively much more common in pears than in apples. In this respect these fruits show the contrary tendency: in the case of pears the groups "poor pollen producers" and "medium pollen producers" contain a comparatively large number of varieties and only about one-fifth of the varieties are "good pollen producers," whereas no less than two-thirds of the apple varieties are "good pollen producers." Thus in apples a high degree of pollen fertility is of common occurrence.

The pollen of such a variety as Gravenstein (lower view in plate 7) is notable not only for its low degree of fertility but also for a considerable amount of variation in the appearance of its pollen grains. There appear plump grains containing granular material and possessing one to four germ pores, plump grain with germ pores but without, or practically without, granular material, plump grains with cytoplasm but without germ pores and, finally grains lacking both granular material and germ pores. If we put the three last-mentioned categories together it is evident even from the appearance of the pollen grains that at least four-fifths of them are not capable of functioning. Fertility tests have further shown that only a small number of the pollen grains containing granular material and one to four germ pores are actually capable of germination.

The tetrads formed through the division of the pollen mother cells frequently contain an abnormal number of microspores. This implies that irregular chromosome distribution has taken place during the meiotic divisions. Shoemaker (1926) has just recently reported that this is the case with the Stayman group, which is marked by great variability in the size of the pollen grains combined with a low degree of fertility. In Gravenstein degeneration of the anther loculi is also often met with, as well as the degeneration in the microspores after their liberation.

#### TESTS OF CROSS-INCOMPATIBILITY IN APPLES

The majority of papers dealing with self-sterility in apples also contain more or less exhaustive tests in regard to cross-incompatibility. As far as America is concerned this is further the case with one paper published by Hedrick and Wellington (1912) and one by Ballard (1916). In Europe Ewert (1922) has recently published some few observations.

The following table showing the Swedish cross-incompatibility tests and comprising the results of 45 crosses has been made up in the same way as the corresponding table in respect to pears.



TABLE 4  
SWEDISH TESTS OF CROSS-INCOMPATIBILITY IN APPLES

SEED PARENT	X	POLLEN PARENT	NUMBER OF BLOSSOMS POLLI- NATED	NUMBER OF BLOSSOMS SET	NUMBER OF FRUITS MATURED	PERCENT- AGE OF BLOSSOMS HAVING PRODUCED MATURE FRUIT
Beauty of Bath	x	Golden Noble.....	230	18	11	4.8
Beauty of Bath	x	Gladstone.....	290	18	18	6.2
Beauty of Bath	x	Bismarck.....	260	38	38	15.4
Belle de Boscoop	x	Cox Pomona.....	192	26	11	5.2
Belle de Boscoop	x	Bismarck.....	110	20	12	10.9
Bismarck	x	Gladstone.....	240	33	30	12.5
Bismarck	x	Grushevka Krasnaja....	270	87	49	17.1
Brunnsäpple of Halland	x	Säfstaholm.....	247	154	33	13.4
Cox Orange	x	Blenheim.....	390	63	54	7.8
Cox Orange	x	Golden Noble.....	145	28	24	16.5
Cox Orange	x	Stenkyrke.....	147	30	29	19.7
Cox Orange	x	King of the Pippins....	115	27	24	20.9
Cox Orange	x	Worcester Pearmain....	88	29	24	27.3
Cox Orange	x	Cox Pomona.....	66	25	23	34.8
Duchess of Oldenburg	x	Bismarck.....	210	73	33	15.7
Gladstone	x	Beauty of Bath.....	200	19	19	9.5
Gladstone	x	Bismarck.....	286	28	28	9.8
Gravenstein	x	Hanaskog.....	235	76	0	0.0
Gravenstein	x	Hörningsholm's Rosenapfel	124	50	4	3.2
Gravenstein	x	Oranie.....	205	44	7	3.4
Gravenstein	x	Kutofsky Nalivia.....	349	64	17	4.9
Gravenstein	x	Langton's Nonsuch.....	439	112	26	5.9
Gravenstein	x	Yellow Transparente....	221	35	16	7.2
Gravenstein	x	Steklianka Kremera....	304	43	24	7.9
Gravenstein	x	P. J. Bergius.....	174	25	15	8.4
Gravenstein	x	Cellini.....	117	21	10	8.5
Gravenstein	x	Arvid.....	364	110	33	9.1
Gravenstein	x	Julity Kutofsky.....	466	118	46	9.8
Gravenstein	x	Säfstaholm.....	441	135	60	13.8
Grushevka Krasnaja	x	Gelber Richard.....	72	14	3	4.2
Grushevka Krasnaja	x	Postophe d'hiver.....	330	118	20	6.1
Grushevka Krasnaja	x	Säfstaholm.....	488	157	63	12.9
Grushevka Krasnaja	x	Bismarck.....	120	24	21	17.5
Ribston	x	Cox Pomona.....	549	2	1	0.2
Stenkyrke	x	Beauty of Bath.....	100	31	7	7.0
Stenkyrke	x	Cox Pomona.....	100	44	9	9.0
Säfstaholm	x	Grushevka Krasnaja....	91	12	6	6.6
Säfstaholm	x	Duchess of Oldenburg....	129	10	9	6.9
Säfstaholm	x	Steklianka Kremera....	237	39	26	10.9
Säfstaholm	x	Kutofsky Nalivia.....	289	46	32	11.1
Säfstaholm	x	Oranie.....	194	49	31	15.9
Yellow Newtown	x	Stenkyrke.....	95	24	14	14.7
Yellow Transparente	x	Bismarck.....	170	25	19	11.2
Yellow Transparente	x	Grushevka Krasnaja....	210	42	34	16.2
Åkerö	x	Yellow Transparente....	178	66	30	17.1

Cross-incompatibility among apples has proved to be of infrequent occurrence in the Swedish tests. Lindfors (1922) mentioned one cross, Gravenstein  $\times$  Hanaskog, in which no fruits at all reached maturity. Whether this is really a case of complete unfruitfulness is somewhat uncertain. Crane (1923) asserts that in renewed tests he always obtained fertilization to the extent of at least some few per cent in such cases. Whether the negative result in Lindfors' test is due to cross-incompatibility is still more uncertain. Since several fruits, representing 4.5 per cent of the number of pollinated flowers, still remained after more than a month from the time of pollination embryo abortion seems more likely to be involved. I have

already pointed out that varieties producing pollen of poor quality often prove to be incapable of competing with "good pollen producers" as pollen parents. But even in those cases where the various pollen parents tested for a certain self-sterile variety all possess pollen of good quality, the results of the pollination experiments may differ considerably from one another. It looks to me, therefore, as if different degrees of cross-compatibility exist in apples. To name now some few extreme cases, the cross Beauty of Bath  $\times$  Golden Noble has produced 4.8 per cent fruits, while the cross Beauty of Bath  $\times$  Bismarck resulted in 15.4 per cent. The cross Gravenstein  $\times$  Hanaskog gave no fruits at all, but the cross Gravenstein  $\times$  Säfstaholm gave 13.8 per cent of mature fruits. The cross Grushevka Krasnaja  $\times$  Gelber Richard gave 4.2 per cent, the cross Grushevka Krasnaja  $\times$  Bismarck, on the other hand, gave 17.5 per cent fruit matured. The cross Säfstaholm  $\times$  Grushevka Krasnaja resulted in 6.6 per cent, but the cross Säfstaholm  $\times$  Oranie gave 15.9 per cent and so on.

The cross Ribston  $\times$  Cox Pomona carried out by E. Johansson (1926) seems to be of some interest. In two consecutive years he obtained extremely poor results. In 1923, 292 blossoms were pollinated with Cox Pomona pollen (which is of very good quality) without any fruits at all being obtained. In 1924 this cross was repeated and 257 blossoms then gave only one fruit. Here then a case of almost complete cross-incompatibility seems to have been found. The author mentions in this connection that Cox Pomona is a seedling of Ribston and probably the explanation lies here.

Cox Orange is another seedling of Ribston. Miss Sutton (1918) crossed these two varieties but obtained only a few fruits. It would be worth while examining these three varieties on a large scale with a view to the phenomenon of cross-incompatibility.

Of great interest from the commercial point of view is the fact that all apple varieties hitherto investigated in Sweden give incomparably larger crops after cross-pollination than when they are restricted exclusively to their own pollen.

#### POLLENIZERS

Which varieties should be selected as pollenizers for commercially important self-unfruitful varieties? This is, from an economic point of view, a very vital question.

A pollenizer should be characterized by an even and profuse bloom and its blooming time should coincide with that of the variety which is to be supplied with foreign pollen. The pollenizer should, further, produce an abundant amount of pollen and its pollen should be of good quality. Those varieties which are to be classified as "poor pollen producers" and which represent no less than 38 per cent of the pear varieties and 14 per cent of the apple varieties investigated in Europe, ought not to be selected as pollenizers for commercially important self-unfruitful varieties. Several of them produce pollen which is viable only to an extent of a few per cent, and further, the few tubes formed in artificial cultures quite often attain only an inconsiderable

length in comparison with the tubes formed by good pollen. Besides, the rate of growth of the pollen tubes is probably often considerably slower in the former than in the latter case.

Through the influence of adverse external factors there is then the risk that the relatively few viable pollen-grains produced by "poor pollen producers" lose, either wholly or in part, their ability to function. As Miss Borgenstam has pointed out, in the case of *Syringa chinensis* a reduction in temperature to zero centigrade results in numerous anomalies in the meiotic divisions, which induce more or less complete sterility. The danger of such frosts is always present, at least in a climate like that of Central Sweden. One should therefore at any rate select a "medium pollen producer" as pollenizer, preferably, of course, a variety characterized by a high percentage of pollen germination. As a rule varieties showing a high degree of pollen viability produce an abundant amount of pollen.

Against this reasoning it has sometimes been objected (N. Johansson, 1923) that there is no very great difference in fertilization if varieties with low, as against varieties with high, power of pollen germination are used as pollenizers. But then the obvious fact is overlooked that artificial pollination is not identical with pollination under natural conditions. In artificial pollination we are very careful to ensure that at least two or three stigmatic surfaces in each flower, and preferably all, show a distinct yellow coloring after the transfer of pollen has taken place. When pollination is effected under such conditions there is actually transferred a considerable quantity of pollen, and it is by no means likely that the flowers would always be so carefully pollinated under natural conditions. Especially if bad weather prevails during the greater part of the flowering period it seems to me generally important that as many insect-visits as possible give positive results. That the power of pollen germination could be greater on the stigmas than in artificial cultures—a further objection that has been brought forward (N. Johansson, 1921)—is an argument which also falls to the ground by reason of its inconsistency in the light of the facts already mentioned regarding the morphology and cytology of pollen. The only way to examine this question further and more thoroughly would be to make counted grain pollinations with such varieties as Alexander Lucas as pollenizers and then to compare the results with those obtained when, for instance, Esperine with an average pollen germination of 82.4 ( $\pm 3.3$ ) per cent, Gansel Bergamot with 85 per cent or Jaminette with 93.5 per cent have been used as pollenizers. No such experiments have been carried out by me as their outcome is self-evident. If we bring down the quantities of pollen low enough, the differences will undoubtedly appear and they will then prove to be very marked. Taking a variety such as Esperine, it is only necessary that two pollen grains become attached to a stigma for fertilization to take place without fail, provided, of course, that cross-compatibility exists, while in the case of a variety such as Marguérite Marillat, whether cultivated in Central Sweden with its rather severe climate for pear-growing or on the plains of Northern Italy, generally



several hundred pollen grains are required to allow of a prospect of even a single one of them being capable of functioning. And perhaps even this quantity will prove insufficient. The extremely few pollen grains of Marguerite Marillat which are capable of germination, i. e., which contain cytoplasm and have formed germ pores, apparently cannot develop normal tubes.

The experiences made by N. Johansson in those of his pollination experiments, in which poor pollen producers were used as pollenizers, are in fact contradicted by the statements of other research-workers. Thus, Kobel (1925) in Switzerland obtained an exactly contrary result. He cross-pollinated the apple varieties Belle de Boscoop, Winterzitronenapfel, Gravenstein and Rheinischer Bohnapfel partly with varieties characterized by poor pollen, viz., Belle de Boscoop, Winterzitronenapfel, Stäfner Rosenapfel and Rheinischer Bohnapfel, and partly with a variety possessing a high percentage of viable pollen, viz., Berner Rosenapfel. The differences in his results were very considerable. With good pollen, i. e., with Berner Rosenapfel as pollenizer, an average fertilization of 36 per cent was obtained, while with poor pollen an average fertilization of only 1 per cent resulted. The two fruits obtained on Rheinischer Bohnapfel after crossing with Belle de Boscoop pollen contained only one plump seed each, the fertilization had consequently been very inadequate. This case is of great interest, since it proves that the poor result of pollination with Belle de Boscoop pollen is not due to physiological cross-incompatibility. It often happened that the fruits formed after pollination with poor pollen soon dropped. Undoubtedly even these fruits contained only a few seeds. E. Johansson (1926) also had discouraging experiences with Belle de Boscoop as a pollenizer. Finally Müller-Thurgau (1908) mentioned the pear variety Beurré Diel as being a bad pollenizer, which agrees with the results of our pollen germination tests.

Close (1902), Auchter (1921 and 1923) and Keil (1923) have found the apple variety Stayman Winesap generally to be very unsatisfactory as a pollen parent in experimental pollination work, and as at the same time the pollen of this variety often does not show a very high power of germination, it is possibly here that the explanation lies. Wellington (1923) has come to the conclusion that, in cross-fertilization tests with apples, both Baldwin and Rhode Island Greening were unsatisfactory pollenizers for some other varieties. This may also at least partly, be due to degeneracy in the male sex. According to Kobel (1925) Baldwin shows in Switzerland a pollen only viable to the extent of 11 per cent and MacDaniels has found in the same variety in the State of New York an average percentage germination of 9 and a maximum one of 13.

From the commercial point of view it seems therefore to be a matter of importance to take account in experimental pollination work of the viability of pollen in different varieties before pollenizers are selected. I do not at all mean, however, that in this way all cases of more or less marked cross-sterility could be explained. Even in pears and apples possibly physiological cross-



incompatibility exists, although occurring far more infrequently in these fruits than in plums and cherries (cf. Crane, 1923).

Finally, it is necessary, from the commercial point of view, to get the fruit as well formed as possible. According to Müller-Thurgau and to Sax (1921) the uniform development of the apple is essentially dependent on seed content. Apples with several adjacent seedless carpels are usually irregular in shape. A similar correlation between seed content and uniform development of the fruit probably exists also in the pear. Thus, in one flower several ovules must be fertilized to enable it to develop into a fruit, which satisfies the grower. This is a further reason for selecting varieties producing an abundant amount of viable pollen as pollenizers. The quantity of fruit and seed and the quality of the fruit improves in a certain degree with the amount of pollen (Correns, 1900).

### TYPES OF STERILITY IN PEARS AND APPLES

In concluding this account of the Swedish pollination tests I shall try to summarize briefly the present position as regards our knowledge of the different types of sterility in pears and apples.

On the lines of the classification made in Stout's memoir on self- and cross-pollinations in *Cichorium* (1916), we seem to have, in pears and apples also, to distinguish between the following main types of sterility:

- I. Sterility from impotence;
- II. Sterility from incompatibility; and
- III. Sterility from embryo abortion.

#### I. STERILITY FROM IMPOTENCE

Leaving aside the question of sterility from non-flowering and abscission due to unfavorable internal nutritive conditions or external factors of various kinds, we have to deal with different degrees of impotence in the male and female sexes. Decidedly one-sided impotence, however, is of very rare occurrence in pears and apples.

Müller-Thurgau (1898) described several years ago a case of total sterility in the male sex. It was an apple variety called Sans Pépins, which was cultivated at a villa in Zürich in Switzerland. The flowers were entirely without stamens but contained 14 to 15 carpels instead of the normal number of five. Each carpel contained two ovules, which were very differently advanced in development at the flowering season; many seemed to be aborted. Although the tree blossomed fairly abundantly each year there were always only a few seedless fruits produced. Evidently there were no other suitable varieties in the neighborhood which could serve as pollen parents.

An entirely contrary case is probably represented by the German apple variety Vaterapfel ohne Kern. In this variety the fruits, according to Müller-Thurgau (1898), contain only in rarely exceptional cases any plump seed, and as a general rule all the seeds are aborted. The pollen, on the other hand, shows a germination power of 95 per cent (Ewert, 1906).

Wallis (1911) mentioned an apple seedling from Australia which al-

though 15 years old never bore any fruit. The flowers were found to be totally unsexual with the carpels entirely lacking.

With regard to pears, Müller-Thurgau (1898) has given an account of Lebrun's Butterbirne and declared that its fruits are constantly lacking in plump seeds. Here, however, even the pollen is of poor quality. Finally, in the pear variety Rihis Kernlose Butterbirne, whose pollen is of medium quality, Ewert (1909) has found the same phenomenon existing.

As to the partial abortion in the female sex the data available are still very meager. Of course there is normally a degeneration of three megaspores in each tetrad, but even in the remaining 25 per cent abortion certainly takes place in several varieties.

Partial sterility from impotence in the male sex exists on a large scale in pears and is also often met with in apples. Pollen fertility and pollen sterility are not to be regarded as contrasted characters as there is no sharp distinction between presence and absence of good pollen, and many grades of pollen abortion occur in both pears and apples. Any one variety appears to be characterized by the production of a certain quantity of pollen with a more or less constant range of abortion and degree of viability. Thus pollen sterility is obviously in the first place an inherent characteristic.

As to the origin of the condition of male sterility in these fruits the paper by Shoemaker (1926) on the development of apple pollen as well as my own preliminary observations on the pollen development in the pear variety Alexander Lucas seem to be of some interest. It has indeed been possible to show the likelihood of hybridization playing an important role in inducing pollen sterility in apples as well as in pears.

Besides pollen sterility as a result of irregular chromosome distribution during the meiotic divisions, there occurs degeneration in whole anther loculi at the pollen mother cell stage as well as in the maturing gametophyte after the tetrad division has been completed.

## II. STERILITY FROM INCOMPATIBILITY

In the case of certain apple varieties one might perhaps speak of morphological incompatibility, since structural differences which render self-pollination difficult occur to a certain extent. Such differences were first observed by Ewert (1906), who found that each pear and apple variety was characterized by its own special flower-structure, which remained constant. In the apple variety Glanzreinette the pistils reach 1 cm. above the anthers, whereby self-pollination is naturally rendered to some degree difficult. A similar state of things was found in the pear variety Elizabeth. In other varieties Ewert found equally long pistils and stamens and in still other varieties pistils which were shorter than the stamens. In the last-mentioned case self-pollination has of course the advantage over cross-pollination. In Sweden Lindfors (1922) has investigated cultivated apple varieties from this point of view and has divided them into 6 groups according to the relative length of stamens and pistils. The most extreme groups, between which a number of transi-

tions exist, were characterized by the anthers reaching somewhat above the pistils and the pistils reaching 8-10 mm. over the stamens, respectively. Nilsson (1924) has called this phenomenon incomplete heterostyly. Besides this there occur as a hindrance to self-pollination differences in the time of development between the anthers and stigmata—more or less marked protogyny—without these differences being in any way connected with the relative length of pistils and stamens (Lindfors, 1922) as was thought by Ewert (1906).

Ewert (1922) determined the number of seeds in the fruits of varieties characterized by long pistils as compared with varieties possessing short pistils, and found that on an average the former contained a greater number of seeds. However, it was not possible to prove any difference in compatibility between the crosses "longistyle"  $\times$  "longistyle" and "brevistyle"  $\times$  "brevistyle" on the one hand and the cross of "longistyle"  $\times$  "brevistyle," or *vice versa*, on the other.

From the investigations carried out both in Sweden and elsewhere it has been shown that complete or partial physiological self-incompatibility very often occurs in cultivated pears and apples. Complete self-compatibility seems to be of very rare occurrence. In wild species of apples, Stout (1925) has, however, found different degrees of self-compatibility ranging from complete self-fertility to complete self-sterility. The existence of at least some wild species of apples showing a high degree of self-compatibility is no doubt of interest to the breeder of new varieties for cultural purposes.

Complete or nearly complete cross-incompatibility from physiological causes, on the other hand, seems to be rare both in pears and apples. In England, Miss Sutton (1918) came to the following conclusion in the case of apples: ". . . provided that a variety produces plenty of pollen and flowers simultaneously with the variety which it is intended to pollinate, any variety . . . will probably serve for this purpose. . . ." Nor have either Rawes (1922) or Crane (1923) observed any clear example of cross-incompatibility in apples. On the other hand Auchter and Schrader (1926) have quoted several cases of cross-sterility which have occurred in investigations carried out in the United States. From the point of view of commercial fruit-growing it is of special interest to have this question definitely decided, which is only possible by means of extended cross-pollination tests carefully carried out.

### III. STERILITY FROM EMBRYO ABORTION

That sterility due to degeneration of embryos during early stages of development also occurs in pears and apples there is no reason to doubt. But what part this type of sterility plays in the dropping of fruit before ripening, as also its relation to the other types, are still but little known. It seems probable, as Stout (1920) has pointed out, that in certain cases embryo abortion is a kind of physiological incompatibility expressing itself after an apparently normal fusion of sex cells. Kraus (1915) thinks that in self-sterile varieties the union of the proper nuclei within the embryo sac generally



takes place and Namikawa (1923) found the growth of pollen tubes in self-pollinated apple flowers to be not in any way abnormal or delayed. Osterwalder (1910) on the contrary is of the opinion that the failure in self-pollination is due to the inability of the pollen tubes to grow deep enough in the styles, and Knight (1918) considers the relatively slow rate of growth of the pollen tubes to be an important factor in self-sterility in the apple.

There is thus some controversy on this point and much work remains to be done here as well as in the case of the other types of sterility in fruit-trees. Our knowledge of the causes of unfruitfulness in pears and apples is, however, consistently increasing from year to year, and the best means of rapidly reaching the goal in this respect I consider to be the international cooperation among geneticists, physiologists and horticulturists, at which this Conference aims.

### BIBLIOGRAPHY

- Adams, J., 1916. On the germination of the pollen grains of apple and other fruit trees. *Bot. Gaz.* **61**: 131-147.
- Alderman, W. H., 1918. Experimental work on self-sterility of the apple. *Proc. Amer. Soc. Hort. Sci.* **1917**: 94-101.
- Andersson, E., 1924. Studies on self-sterility. The genetic basis of cross-sterility in *Nicotiana*. *Genetics* **9**: 13-40.
- Auchter, E. C., 1922. Apple pollen and pollination studies in Maryland. *Proc. Amer. Soc. Hort. Sci.* **1921**: 51-80.
- Auchter, E. C., 1923. Importance of proper pollination in fruit yields. *Peninsula (Md. and Del.) Hort. Sci.*
- Auchter, E. C., and Schrader, A. L., 1926. Cross-fertilization of the Arkansas (Mammoth Black Twig) apple. *Proc. Amer. Soc. Hort. Sci.* **1925**: 96-105.
- Ballard, W. R., 1916. Methods and problems in pear and apple breeding. *Maryland Agric. Exp. Sta. Bull.* **196**.
- Backhouse, W. O., 1912. The pollination of fruit trees. *The Gardeners' Chronicle* **52**: 381.
- Beach, S. A., Booth, N. O., and Taylor, O. M., 1905. The apples of New York. *Rept. New York Agric. Exp. Sta. for 1903*.
- Beaumont, J. H., and Knight, L. I., 1923. Apple pollen germination studies. *Proc. Amer. Soc. Hort. Sci.* **1922**: 161-163.
- Borgenstam, E., 1922. Zur Zytologie der Gattung *Syringa* nebst Erörterungen über den Einfluss äusserer Faktoren auf die Kernteilungsvorgänge. *Ark. f. Bot. (K. Svenska Vetenskapsakademien)*. **17**: No. 15. 1-27.
- Chandler, W. H., 1925. *Fruit Growing*.
- Chittenden, F. J., 1914. Pollination in orchards. III. Self-fruitfulness and self-sterility in apples. *Jour. Roy. Hort. Soc.* **39**: 615-628.
- Close, C. P., 1903. Pollination of pears, peaches and apples. *Del. Agric. Exp. Sta. Rept.* **14**: 99-102.
- Correns, C., 1900. Ueber den Einfluss welchen die Zahl der zur Bestäubung verwendeten Pollenkörner auf die Nachkommenschaft hat. *Ber. Deutsch. Bot. Ges.* **18**: 422-435.
- 1912. Selbsterilität und Individualstoffe. *Biol. Centralbl.* **33**: 389-423.
- Crandall, C. S., 1922. Results from self-pollination of apple flowers. *Proc. Amer. Soc. Hort. Sci.* **1921**: 95-100.
- Crane, M. B., 1923. Report on tests of self-sterility and cross-incompatibility in plums, cherries and apples at the John Innes Horticultural Institution. II. *Jour. of Pomol. and Hort. Sci.* **3**: 67-84.
- Dahl, C. G., and Sonesson, N., 1913. Undersökningar rörande tiderna för fruktträdens blomning under åren 1910-1912. *Sveriges Pomol. Fören. Årsskr.* **14**: 135-147.
- Davis, B. M., 1923. Pollen- and seed-sterility in hybrids. *Amer. Jour. Bot.* **10**: 462-467.
- Dorsey, M. J., 1919. A study of the sterility in the plum. *Genetics* **4**: 417-486.
- 1922. The set of fruit in apple crosses. *Proc. Amer. Soc. Hort. Sci.* **1921**: 82-94.
- 1923. Sterility in relation to horticulture. *Amer. Jour. Bot.* **10**: 474-484.
- East, E. M., 1915. An interpretation of self-sterility. *Proc. Nat. Acad. Sci.* **1**: 95-100.
- East, E. M., and Mangelsdorf, A. J., 1925. A new interpretation of the hereditary behavior of self-sterile plants. *Proc. Nat. Acad. Sci.* **11**: 166-171.



- East, E. M., and Park, J. B., 1918. Studies on self-sterile plants. II. Pollen tube growth. *Genetics* 3: 353-366.
- Ernst, A., 1918. Bastardierung als Ursache der Apogamie im Pflanzenreich.
- Ewert, R., 1906. Blütenbiologie und Tragbarkeit der Obstbäume. *Landw. Jahrb.* 35: 259-287.
- 1909. Neuere Untersuchungen über Parthenokarpie bei Obstbäumen und einigen anderen fruchttragenden Gewächsen. *Landw. Jahrb.* 38: 767-839.
- 1910. Die korrelativen Einflüsse des Kerns beim Reifeprozess der Früchte. *Landw. Jahrb.* 39: 471-486.
- 1921. Förderung der Fruchtbarkeit der Obstbäume durch Bienenzucht. *Landw. Jahrb.* 56: 74-82.
- 1922. Förderung der Fruchtbarkeit der Obstbäume durch Bienenzucht. *Landw. Jahrb.* 57: 76-79.
- Fletcher, S. W., 1900. Pollination in orchards. *Cornell Agric. Exp. Sta. Bull.* 181.
- 1911. Pollination of Bartlett and Kieffer pears. *Virg. Agric. Exp. Sta. Ann. Rpt. for 1909-10*, pp. 213-224.
- Florin, E. H., 1925. Pollinering och fruktsättning hos päronsorster. (Pollination of pears.) *Medd. Perm. Komm. f. Fruktodlingsförsök. (Contrib. Swedish Perm. Comm. on Orchard Research.)* No. 5. Stockholm. Pp. 1-39.
- 1926. Germination tests with pollen of apples and pears, 1923-1925. Unpublished.
- 1926. Pollination experiments on apples and pears, 1923-1926. Unpublished.
- Florin, R., 1920. Zur Kenntnis der Fertilität und partiellen Sterilität des Pollens bei Apfel- und Birnensorten. *Acta Horti Bergiani* 7: 1-39.
- 1921. Biologiska undersökningar av fruktträd IV. *Sveriges Pomol. Fören. Årsskr.* 22: 1-13.
- 1926. Sambandet mellan pollinering och fruktsättning hos fruktträdsorster. *Svenska Lantbruksveckans Helsingfors Handlingar*, 6: 118-156.
- Gardner, V. R., Bradford, F. C., and Hooker, Jr., H. D., 1922. The fundamentals of fruit production.
- Gowen, J. W., 1920. Self-sterility and cross-sterility in the apple. *Maine Agric. Exp. Sta. Bull.* 287.
- Hedrick, U. P., 1921. The pears of New York. *New York State Agric. Exper. Sta. Rpt.*
- Hedrick, U. P., and Wellington, R., 1912. An experiment in breeding apples. *New York State Agric. Exp. Sta. Bull.* 350.
- Heinicke, A. J., 1917. Factors influencing the abscission of flowers and partially developed fruits in the apple (*Pyrus Malus* L.). *Cornell Agric. Exp. Sta. Bull.* 393.
- Hooper, C. H., 1921. Pollination of fruits. *Jour. Ministry of Agric.* 28: 124-133.
- Johansson, E., 1926. Blombiologiska försök vid Alnarp 1923-1925. (Pollination experiments and studies of pollen quality at Alnarp.) *Sveriges Pomol. Fören. Årsskr.* 27: 1-30.
- Johansson, N., 1921. Blombiologiska försök å fruktträd 1921. *Sver. Pomol. Foren. Årsskr.* 22: 147-153.
- 1923. Pollinerings- och kombinationsförsök med fruktträd. *Sver. Pomol. Fören. Årsskr.* 24: 17-28.
- Keil, J. M., 1923. Apple pollination. *Ohio Agric. Exper. Sta. Bull.* 8.
- Kihara, H., 1924. Cytologische und genetische Studien bei wichtigen Getreidearten mit besonderer Rücksicht auf das Verhalten der Chromosomen und die Sterilität in dem Bastarden. *Mem. Coll. of Sci. Kyoto Imp. Univ., Ser. B.* 1: 1-200.
- Knight, L. J., 1918. Physiological aspects of self-sterility of the apple. *Proc. Amer. Soc. Hort. Sci.* 1917: 101-105.
- Knowlton, H. E., 1921. Methods in apple-pollination experiments. *Proc. Amer. Soc. Hort. Sci.* 1920: 44-47.
- and Sevy, H. P., 1926. The relation of temperature to pollen tube growth in vitro. *Proc. Amer. Soc. Hort. Sci.* 1925: 110-115.
- Kobel, F., 1925. Die Keimfähigkeit des Pollens einiger wichtiger Apfel- und Birnensorten und die Frage der gegenseitigen Befruchtungsfähigkeit dieser Sorten. *Landw. Jahrb. d. Schweiz.* 38: 443-461.
- Kraus, E. J., 1915. The self-sterility problem. *Jour. of Heredity* 6: 549-557.
- Kroemer, K., und Gessmann, E., 1924. Untersuchungen über die Keimfähigkeit des Pollens bei Obstbäumen im Frühjahr. 1923. *Landw. Jahrb.* 60: 487-489.
- Lewis, C. I., and Vincent, C. C., 1909. Pollination of the apple. *Ore. Agric. Exp. Sta. Bull.* 104.
- Lindfors, Th., 1922. Om pollination och fruktsättning hos *Gravensteiner* och *Akerö*. *Sveriges Pomol. Fören. Årsskr.* 23: 172-176.

- 1922. Bidrag till våra äpplesorters blombiologi. Sveriges Pomol. Fören. Arsskr. 23: 271-276.
- Lindhard, E., 1919. Aebleblomstens befrugtning. Tidskr. f. Planteavl. 26: 538-542.
- Ljungdahl, H., 1921-1922. Zur Zytologie der Gattung *Papaver*. Svensk Bot. Tidskr. 16: 103-114.
- Logsdail, A. J., 1917. Rept. Plant Breeding. Canada Experimental Farms Rept. 1915-1916.
- Macoun, W. T., 1923. Preliminary report on self-pollination studies. Report of the Dominion Horticulturist for the year 1922. Canada Dept. of Agric., pp. 13-15.
- MacDaniels, L. H., 1926. Pollination studies with certain New York State apple varieties. Proc. Amer. Soc. Hort. Sci. 1925: 87-96.
- Manaresi, A., 1912. Ricerche sul polline degli alberi fruttiferi. Le Staz. Sperim. Agrar. Ital. 45: 809-873.
- Martin, J. N., and Yocum, L. E., 1918. A study of the pollen and pistils of apples in relation to the germination of the pollen. Proc. Iowa Acad. Sci. 25: 391-410.
- Michaelis, P., 1926. Ueber den Einfluss der Kälte auf die Reduktionsteilung von *Epilobium*. Planta 1: 569-582.
- Middlebrooke, W. J., 1915. Pollination of fruit trees. Jour. Board of Agric., Canada 22: 418-433.
- De Mol, W., 1923. Duplication of generative nuclei by means of physiological stimuli and its significance. Genetica 5: 225-272.
- Morris, O. M., 1921. Studies in apple pollination. Wash. Agric. Exp. Sta. Bull. 163.
- Müller-Thurgau, H., 1898. Abhängigkeit der Ausbildung der Traubenbeeren und einiger anderer Früchte von der Entwicklung der Samen. Landw. Jahrb. d. Schweiz 12: 135-202.
- 1903. Die Folgen der Bestäubung bei Obst- und Rebenblüten. Ber. d. Zürich. Bot. Ges. 13: 45-63.
- 1905. Die Befruchtungsverhältnisse bei den Obstbäumen. Landw. Jahrb. d. Schweiz. 19: 564-566.
- 1908. Kernlose Traubenbeeren und Obstfrüchte. Landw. Jahrb. d. Schweiz. 22: 560-593.
- 1908. Weitere Untersuchungen über die Befruchtungsverhältnisse bei den Obstbäumen. Landw. Jahrb. d. Schweiz. 22: 755-758.
- Namikawa, I., 1922. Ueber die vorzeitige Abstossung der jungen Früchte von *Malus communis*. Jour. Coll. Agric. Hokkaido Imp. Univ. 11: 1-21.
- 1923. Growth of pollen tubes in self-pollinated apple flowers. Bot. Gaz. 76: 302-310.
- Nilsson, E., 1924. Iakttagelser öfver några blom morfologiska egenskaper hos *Anchusa officinalis* L. och deras variation. Bot. Not. Lund., pp. 393-409.
- van Oijen-Goethals, M. C., 1912-13. Bestuiving en vruchtbaarheid van Ooftboomen. Maandl. d. Nederl. Pom. Ver., pp. 3-43.
- 1916-17. Verslag der Bestuivingproeven over 1916. Maandbl. d. Nederl. Pom. Ver., pp. 164-176.
- Osterwalder A., 1907. Untersuchungen über das Abwerfen junger Kernobstfrüchte. Landw. Jahrb. d. Schweiz 21: 215-224.
- 1909. Ueber das Abwerfen der Blüten unserer Kernobstbäume. Landw. Jahrb. d. Schweiz 23: 339-350.
- 1910. Blütenbiologie, Embryologie und Entwicklung der Frucht unserer Kernobstbäume. Landw. Jahrb. 39: 917-998.
- Overholser, E. L., 1919. Pollination of apples. Rept. Coll. of Agric. and Agric. Exp. Sta., Univ. of Calif., 1918-1919, pp. 28-29.
- Overholser, E. L., and Cameron, 1922. Pollination of apples. Rept. Coll. of Agric. and Agric. Exp. Sta., Univ. of Calif., 1921-1922, pp. 127-128.
- Overholser, E. L., and Jacob, H. E., 1921. Pollination of apples. In Rept. Coll. of Agric. and Agric. Exp. Sta., Univ. of Calif., 1920-1921, p. 54.
- Passecker, F., 1926. Untersuchungen über die Fertilität des Pollens verschiedener Obstsorten. Fortschritte der Landwirtschaft. 1: 46-48.
- Powell, G. H., 1901. Kieffer pear pollinations. The pollination of apples. Ann. Rept. Del. Agric. Exp. Sta. 13: 129-139.
- 1902. Report of the horticulturist. Ann. Rep. Del. Agric. Exp. Sta. 13: 83-126.
- Prescott, E. E., 1911. Pollination of pear blossoms. Jour. of Agric., Victoria 9: 1-10.
- Rawes, A. N., 1922. Pollination in orchards (V). Summary of apple pollination investigations. Jour. Roy. Hort. Soc. 47: 8-14.
- Rosenberg, O., 1917. Die Reduktionsteilung und ihre Degeneration in *Hieracium*. Svensk Bot. Tidskr. 11: 10-206.

- 1926. Zum Mechanismus der diploiden Kernteilung. Arkiv. f. Bot. (K. Svenska Vetenskapsakademien.) 20: B. No. 3, pp. 1-5.
- Sakamura, T., 1920. Experimentelle Studien über die Zell- und Kernteilung. Jour. Coll. Sci. Imp. Univ. Tokyo 39: 1-221.
- Sax, K., 1921. Studies in orchard management. II. Factors influencing fruit development of the apple. Maine Agric. Exp. Sta. Bull. 298.
- 1922. Sterility relationships in Maine apple varieties. Maine Agric. Exp. Sta. Bull. 307.
- Shoemaker, J. S., 1926. Pollen development in the apple, with special reference to chromosome behavior. Bot. Gaz. 81: 148-172.
- Sonesson, N. 1914. Iakttagelser över fruktträdens blommingsbiologi. Sveriges Pomol. Fören. Årsskr. 15: 141-160.
- Stout, A. B., 1916. Self- and cross-pollinations in *Cichorium Intybus* with reference to sterility. Mem. New York Bot. Gard. 6: 333-454.
- 1918. Fertility in *Cichorium Intybus*: self-compatibility and self-incompatibility among the offspring of self-fertile lines of descent. Jour. Genetics 7: 71-103.
- 1920. Further experimental studies on self-incompatibility in hermaphrodite plants. Jour. Genetics 9: 85-129.
- 1923. The physiology of incompatibilities. Am. Jour. Bot. 10: 459-461.
- 1925. Self-incompatibility in wild species of apples. Jour. New York Bot. Gard. 26: 25-31.
- Stålfelt, M. G., 1920. Sveriges Pomologiska Förenings pollineringsundersökningar, 1919. Sveriges Pomol. Fören. Årsskr. 21: 26-33.
- 1921. Självfertilitet, självsterilitet och partenokarpi hos vara fruktsorter. Sveriges Pomol. Fören. Årsskr. 22: 52-55.
- Sutton, Ida, 1918. Report on tests of self-sterility in plums, cherries and apples at the John Innes Horticultural Institution. Jour. Genetics 7: 281-300.
- Tischler, G., 1908. Zellstudien an sterilen Bastardpflanzen. Arch. f. Zellforsch. 1: 33-151.
- 1912. Ueber die Entwicklung der Samenanlagen in parthenokarpen Angiospermen-Früchten. Jahrb. f. wiss. Bot. 52: 1-84.
- 1921-22. Allgemeine Pflanzenkaryologie. In Linsbauer's Handbuch der Pflanzenanatomie. Allgemeiner Teil: Cytologie. Bd. 2, pp. 1-899.
- Tufts, W. P., 1919. Pollination of the *Bartlett* pear. Calif. Agr. Exp. Sta. Bull. 307.
- and Philp, G. L., 1923. Pear pollination. Calif. Agr. Exp. Sta. Bull. 375.
- Täckholm, G., 1922. Zytologische Studien über die Gattung *Rosa*. Acta Horti Bergiani 7: 97-381.
- Waite, M. B., 1894. The pollination of pear flowers. U. S. Dept. of Agric., Div. of Veget. Pathol. Bull. 5.
- 1899. The pollination of pomaceous fruits. Yearb. U. S. Dept. of Agric. (1898), pp. 167-180.
- Wallis, E., 1911. Sterility in fruit trees. Jour. Dept. Agric. Victoria 9: 10-19.
- Wellington R., 1923. Self-sterility and self-fertility of fruit-varieties grown in New York. New York State Agr. Exp. Sta. Circ. No. 71.
- 1924. An experiment in breeding apples. II. New York State Agric. Exp. Sta. Techn. Bull. 106.
- Wicks, W. H., 1918. The effect of cross-pollination on size, color, shape and quality of the apple. Arkansas Agr. Exp. Sta. Bull. 143.
- Zacharias, E., 1911. Ueber Frucht- und Samenanlage von Kulturpflanzen. Zschr. f. Bot. 3: 785-795.

## ADDENDUM

Since this account was already written and delivered for publication the following papers dealing with the same subject have reached me but unfortunately too late to be considered in this paper.

- Asami, Y., 1926. Preliminary report on the self-sterility of Japanese pear. Chojuro. Proc. Imp. Acad. Tokyo 2: 139-141.
- Evert, R., 1926. Pflanzenphysiologische und biologische Forschungen im Obstbau. Landw. Jahrb. 64: 759-785.
- Kawecka, B., 1926. Études sur le pollen des poiriers et des pommiers. Bull. de l'Acad. Polon. des Sci. et des Lettr., Cl. des Sci. Math. et Nat., Sér. B (1925). Pp. 847-876.
- Kobel, F., 1926. Untersuchungen über den Fruchtsatz unserer Obstarten. Schweizer. Zschr. f. Obst- u. Weinbau in Wädenswil 1926: 1-12.



- 1926. Die zytologischen Ursachen der partiellen Pollensterilität bei Apfel- und Birnsorten. Archiv d. Julius Klaus-Stiftung f. Vererbungsforschung, Sozialanthropologie u. Rassenhygiene 2: 39-57.
- 1926. Ursachen und Folgen der teilweisen Pollensterilität verschiedener Apfel- und Birnsorten. Landw. Jahrb. d. Schweiz. 1926: 441-462.
- 1926. Untersuchungen über die Keimfähigkeit des Pollens unserer wichtigsten Stein- und Kernobstsorten mit einem Überblick über die Befruchtungsverhältnisse derselben. Landw. Jahrb. d. Schweiz. 1926: 550-589.
- Pashkewitsh, V. V., 1925. Relation of the number of flowers on the fruit trees to that of the fruits setting and of the fruits matured. Bull. Applied Bot. and Plant Breeding 14: 119-130.
- 1925. Influence of proper and alien pollen of different varieties on the forming and the maturing of the apple-fruit. Bull. Applied Bot. and Plant Breeding 14: 91-103.
- Petrov, A. V., 1925. Experiments on the influence of self-pollination and cross-pollination on the forming and the variation of the apple-fruit. Bull. Applied Bot. and Plant Breeding 14: 104-118.
- Rybin, V. A., 1926. Cytological investigations of the genus *Malus*. Bull. Applied Bot. and Plant Breeding 16: 187-200.
- Ziegler, A., and Branscheidt, P., 1927. Pollenphysiologische Untersuchungen an Kern- und Steinobstsorten in Bayern und ihre Bedeutung für den Obstbau. Berlin.

## EXPLANATION OF PLATES

### PLATE 5

- Typical germinations of pollen of pears in 10% cane sugar solution after 24 hours.
1. Pollen of the variety Alexander Lucas, one of the "poor pollen producers." Mean percentage of germination is  $2.1 (\pm 1.2)$ .
  2. Pollen of the variety Bartlett, one of the "medium pollen producers." Mean percentage of germination is  $53.8 (\pm 3.0)$ .
  3. Pollen of the variety Hofsta, one of the "good pollen producers." Mean percentage of germination is  $78.0 (\pm 2.4)$ .

### PLATE 6

- Stages in the development of pollen in the pear variety Alexander Lucas, showing much pollen abortion.
- 1 and 2. The two sections of a pollen mother cell in diakinesis showing bivalents and univalents.
  3. Early metaphase with multipolar spindle.
  4. Late anaphase with delayed univalents.
  5. Normal interkinesis; all chromosomes assembled in two nuclei of nearly equal size.
  6. Interkinetic nuclei of unequal size having acquired a different number of chromosomes.
  7. Telophase showing some chromosomes which had not reached the poles before the membrane of the two daughter nuclei was developed.
  8. Showing two main and five micronuclei, the latter resulting from delayed chromosomes.
  9. Interkinesis with one micronucleus at the side of the spindle.
  10. Homoeotypic division showing two larger and two smaller spindles.
  11. Six microspores within the "tetrad" wall, of which two are very small.
  12. Group of pollen grains illustrating various kinds of abortion.

### PLATE 7

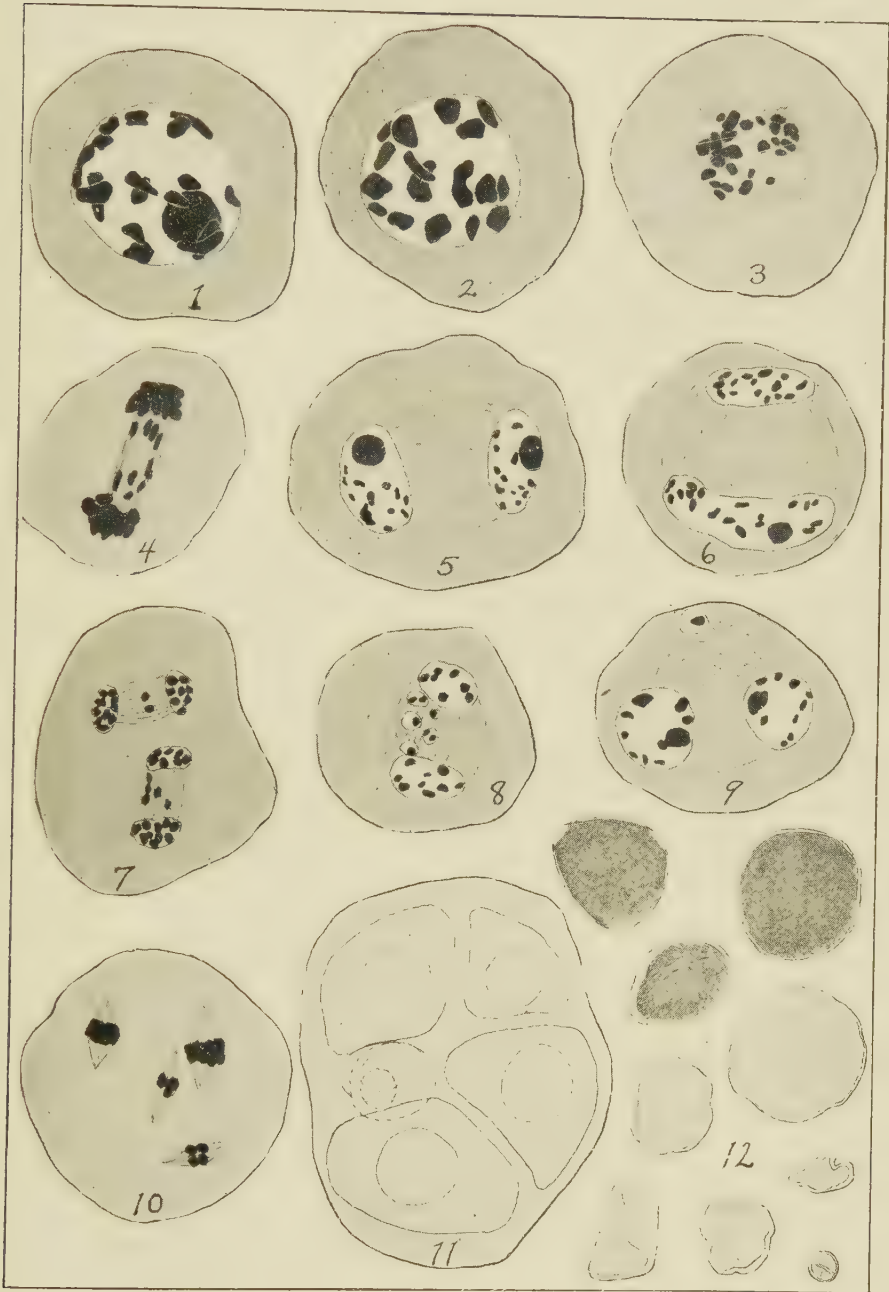
- Typical germinations of pollen of apples in 10% cane sugar solution after 24 hours.
1. Pollen of the variety Gravenstein, one of the poorest of the "poor pollen producers." Mean percentage of germination is  $7.3 (\pm 1.3)$ .
  2. Pollen of the variety Springgrove Codlin, one of the "medium pollen producers." Mean percentage of germination is 45.5.
  3. Pollen of the variety Lord Suffield, one of the "good pollen producers." Mean percentage of germination is  $94.6 (\pm 3.6)$ .





FLORIN: POLLEN PRODUCTION

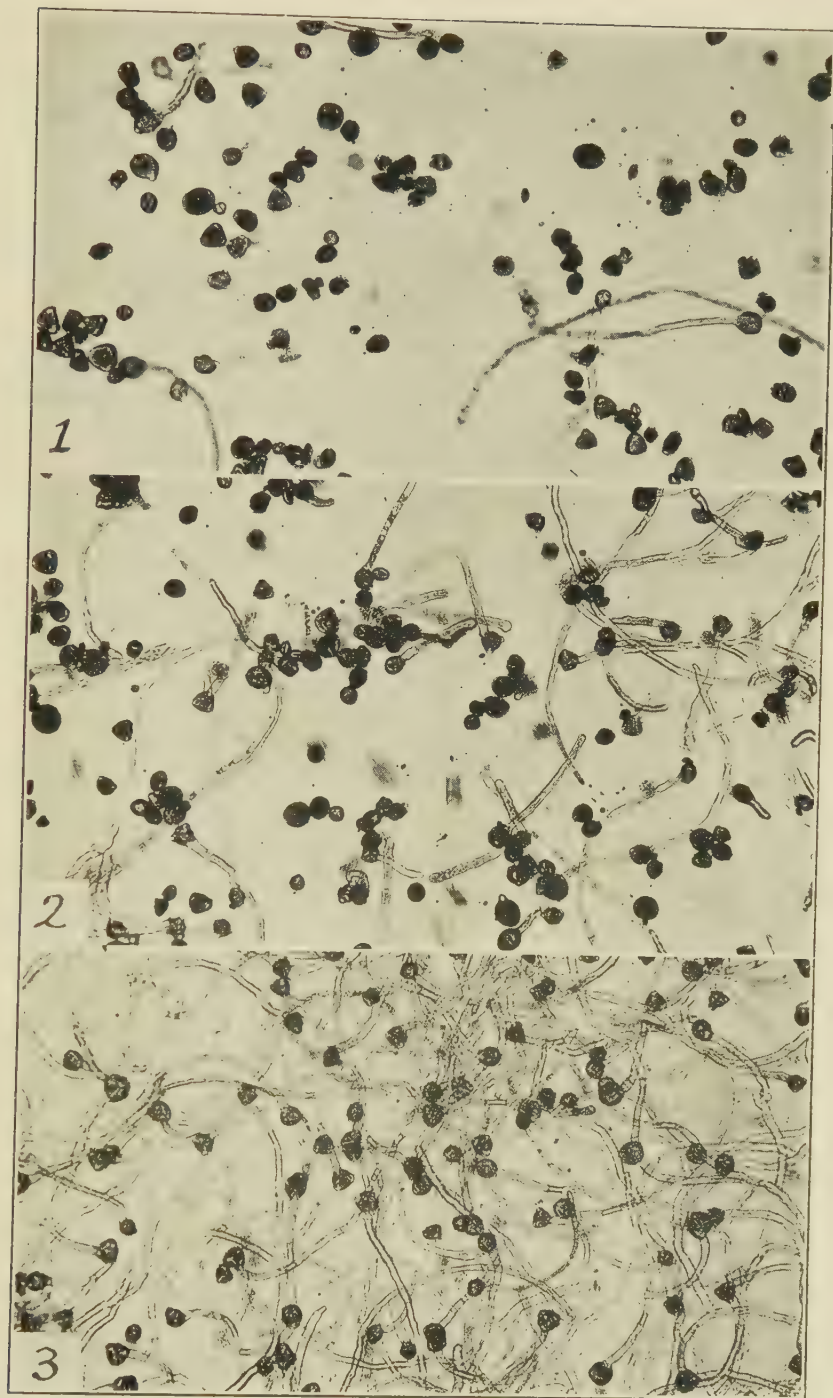




FLORIN: POLLEN PRODUCTION







FLORIN: POLLEN PRODUCTION



# STUDIES IN RELATION TO STERILITY IN PLUMS, CHERRIES, APPLES AND RASPBERRIES

M. B. CRANE

*The John Innes Horticultural Institution, Merton, England*

(WITH PLATES 8-12)

That this Conference is devoted to the problems associated with "Flower and Fruit Sterility," is, I think, a matter for congratulation. It indicates that, apart from its scientific aspects, the horticultural importance of these phenomena is widely recognized.

The phase of the general subject upon which I have to report relates to sterility in fruit trees, and I shall first attempt to give a summary of the more practical results of the experiments carried out at the John Innes Horticultural Institution, and secondly a brief account of related genetical and cytological observations.

In early pomological literature we occasionally find records of fruit trees failing to bear satisfactory crops, which, in the light of present day knowledge, leave but little doubt that self-incompatibility was involved. It was not, however, until the work of Waite (1894) in this country that the phenomenon of self-sterility or self-incompatibility in fruit trees was adequately demonstrated and its economic importance realized.

Since then this subject has received the attention of numerous workers in many countries, and it is beyond the scope of this report to attempt an historical survey of the progress made and adequately to acknowledge the important results they have achieved.

Our experiments began in 1911, and have been continued in each succeeding year. At the commencement flowers were isolated with paper bags in the open, but the effects of frost, cold winds, heavy rains, etc., frequently impaired the accuracy of the results, and from 1912 all the experiments have been carried out on trees grown in pots in an orchard house especially designed for the purpose of these experiments. This house is provided with mosquito-bars and various other precautions are taken to exclude insects. The provision of these facilities not only eliminates many disturbances but affords greater convenience in regard to the manipulations and the results are more reliable.

## CHERRIES

### SELF-INCOMPATIBILITY

We have so far tested thirty-three varieties of the sweet cherry. The majority of these wholly fail to set fruit with their own pollen, and for practical purposes the whole may be regarded as self-incompatible, as from a

total of 25,840 flowers self-pollinated only twenty-three fruits have formed and reached maturity. It may be recalled that the work of others, e. g., Gardner (1913) and Schuster (1922, 1925) in this country, Florin (1923) in Sweden, Sprenger (1908) in Holland and Hooper (1924) in England, also show that it is exceptional in this group to obtain fruits from self-pollinated flowers. Therefore, at least from a grower's point of view, self-incompatibility appears to be universal in the sweet cherry.

#### CROSS-INCOMPATIBILITY

Among varieties of the sweet cherry, we have met with several examples of cross-incompatibility, and as far as our work has progressed we have found that the varieties involved in this inter-sterility can be assigned to groups within which all self and cross-pollinations fail. It is, however, noteworthy that Gardner (1913) has reported an example of cross-incompatibility in the sweet cherry which is effective in one way of a cross but fails in the reciprocal.

So far we have found thirteen varieties which are involved in cross-incompatibility. The varieties and the intra-incompatible groups in which they occur are as follows:

#### GROUP I

Early Rivers  
Bedford Prolific  
Black Tartarian  
Black Tartarian "A"  
Knight's Early Black  
Black Eagle  
Roundell

#### GROUP II

Big. de Schrecken  
Big. Frogmore  
Guigne de Winkler  
Waterloo

#### GROUP III

Big. Napoleon  
Emperor Francis

The total results of the pollinations made between these varieties are summarized in TABLE 1. The details have been given more precisely in former reports (Crane, 1923 and 1925).

There are reasons for suspecting that some of the fruits that have formed within these incompatible groups are the result of errors, but some have occurred under very stringent conditions. It is, however, obvious that for all practical purposes incompatibility may be regarded as complete within these groups.

The large majority of the compatible cross-pollinations have resulted in full crops of fruit, thereby proving economically efficient. There are, however, real indications that such matings are not always completely compatible, as some have consistently given comparatively poor results.

Among the sweet cherries varying amounts of bad pollen grains occur, but in no variety has the proportion aborted been large enough to detract from its value as a pollinizer, nor in the practical work of crossing has such a case been met with. Defective seeds are also common in fruits arising from cross-pollinations. They occur more frequently in some combinations than others.

The varieties within these incompatible groups commonly show much diversity, but some of the varieties used in these experiments are very like



TABLE 1

SHOWING SELF AND CROSS COMBINATIONS MADE BETWEEN INCOMPATIBLE VARIETIES OF CHERRIES. THE NUMBERS AT THE TOP OF THE SQUARES ARE THE NUMBER OF FLOWERS POLLINATED, THOSE AT THE BOTTOM ARE THE NUMBER OF FRUITS WHICH SET AND MATURED.

	Bedford Prolific	Black Eagle	Black Tartarian 'A'	Black Tartarian 'B'	Early Rivers	Knight's Early Black	Roundel	Big Frogmore	Big de Schrecken	Guigne de Winkler	Waterloo	Big Napoleon	Emperor Francis
Bedford Prolific	1601 0	195 0	355 0	87 0	637 0	354 0	242 0	511 192	262 51	237 57	273 96	283 77	21 4
Black Eagle	190 0	707 0	134 0		41 0	111 0		45 9	29 3		92 6	41 10	96 7
Black Tartarian 'A'	65 0	231 0	1640 0	42 0	797 0	31 0		311 21	33 4	509 27	34 5	332 26	206 17
Black Tartarian 'B'	119 0	24 0	40 0	403 0	373 0	57 0		118 23		24 6	49 12	145 17	20 4
Early Rivers	558 0	23 0	740 0	481 0	1584 0	597 0	97 0	475 43	280 40	285 26	25 14	62 5	
Knight's Early Black	379 0	256 1	32 0	141 0	378 1	1156 1		99 20	100 27	112 10		161 46	
Roundel	94 0						33 0			36 8			
Big Frogmore	129 59	27 8	17 5	56 10	454 138	452 141		1259 0	1154 7	281 0	211 0	82 13	362 88
Big de Schrecken	674 198	179 46	133 31	65 12	514 139	195 59	75 22	2102 1	2580 0	668 3	170 3	472 117	351 57
Guigne de Winkler	248 123	37 5	32 17	70 12	51 9	90 16	18 8	678 0	414 0	534 0	347 0	131 64	101 41
Waterloo								113 0	93 0	63 0	728 0		
Big Napoleon	114 56		50 32		74 41	92 41	36 19	38 15	117 65	139 56		576 2	1015 0
Emperor Francis	304 100	104 14	31 6	20 11	205 72	33 11	159 32	128 54	47 21	189 73	40 8	957 4	1521 8

one another; owing to this and the fact that considerable confusion prevails in the nomenclature of cherries many difficulties occur. I have, however, endeavored to ensure that the varieties used in these experiments are correctly named. There is no question of their being distinct individuals, but the identity of some remains obscure. I have referred to some of the difficulties met with in the identification of these horticultural varieties in previous reports.

In the sour cherries, *Prunus cerasus* and the Dukes, our work has not been so extensive, but among them varying degrees of self-incompatibility occur, ranging from varieties which wholly fail to others which set a full crop with their own pollen. Crosses between these groups have varied in their ability to form fruits. Sweet varieties as females, pollinated by Duke's and sour varieties as males, generally produce and mature fruits freely, but from reciprocal pollinations fruits are less freely formed. The total results of such crosses are:

Bigarreus and Guignes	x	Morellos, Kentish and Dukes	Morellos Kentish and Dukes	x	Bigarreus and Guignes
Flowers.....		2,347		Flowers.....	1,521
Fruit .....		774=32%		Fruit .....	115=7.5%

These results may be of importance in regard to the varieties that have been used in the experiments, but exceptions have occasionally occurred and, pending further evidence and confirmation from work with more varieties the above results cannot be regarded as implying any general rule.

The crosses between these groups have also varied in their ability to form good seeds and in certain combinations they have been extremely rare, although well developed fruits have formed comparatively freely. Others have given a large proportion of good seeds and from certain of these a number of seedlings have been raised.

The Dukes are generally regarded by systematic pomologists as hybrids between *P. avium* and *P. cerasus*, mainly on the evidence of intermediate characters. In addition the occurrence of a proportion of bad pollen together with defective seeds is sometimes advanced as a sign of their hybridity. It is, however, of interest to recall that in varieties regarded as within *P. avium*, aborted pollen and defective seeds are not uncommon.

Our knowledge regarding the origin of cherry varieties is scanty and but little value would arise from a lengthy discussion. Certain varieties raised by Thomas Andrew Knight (1822) are, however, of particular interest, e. g., Knight's Early Black, Black Eagle and Waterloo were raised from crossing a Bigarreau with May Duke. This shows that the origin of these varieties is complex, although to the eye Black Eagle and Knight's Early Black pass as sweet cherries, i. e., *avium*.

The following gives the classes of the varieties of cherries grouped according to self-compatibility and self-incompatibility with the data of the tests.

#### CHERRIES THAT ARE SELF-COMPATIBLE

Variety	Number of Flowers Selfed	Fruit Set	% Set
Late Duke.....	2,628	264	10.0
Flemish Red .....	84	12	14.2
Wye Morello .....	360	52	14.4
Kentish Red "A" .....	2,044	464	22.7
Morello.....	3,302	936	28.3

#### CHERRIES THAT ARE PARTIALLY SELF-COMPATIBLE

Variety	Number of Flowers Selfed	Fruit Set	% Set
May Duke.....	4,432	53	1.19
Royal Duke.....	1,047	13	1.24
Arch Duke.....	794	17	2.14
Empress Eugene.....	700	24	3.42

## CHERRIES THAT ARE SELF-INCOMPATIBLE

Variety	Number of Flowers Selfed	Fruit Set	% Set
Amber Heart.....	642	0	..
Bedford Prolific.....	1,601	0	..
Belle d'Orleans.....	345	0	..
Belle de St. Trone.....	305	0	..
Big. de Schrecken.....	2,580	0	..
Big. Frogmore.....	1,259	0	..
Big. Jaboulay.....	591	0	..
Big. Late Black.....	1,139	2	.1
Big. Noir de Guben.....	1,425	1	.07
Big. Noir de Schmidt.....	1,017	2	.01
Big. Napoleon.....	576	2	.3
Black Eagle.....	707	0	..
Black Heart.....	94	0	..
Black Tartarian "A".....	1,640	0	..
Black Tartarian "B".....	403	0	..
Black Tartarian "E".....	328	0	..
Bohemian Black.....	626	2	.3
Early Purple Gean.....	249	0	..
Early Rivers.....	1,584	0	..
Elton.....	420	0	..
Emperor Francis.....	1,521	8	.5
Florence.....	105	0	..
Geante d'Hedelfingen.....	188	0	..
Governor Wood.....	1,436	3	.2
Guigne d'Annonay.....	551	0	..
Guigne de Winkler.....	534	0	..
Knight's Early Black.....	1,156	1	.08
Monstreuse de Mezel.....	259	0	..
Noble.....	754	1	.1
Roundell.....	33	0	..
Turkey Heart.....	1,059	1	.09
Waterloo.....	728	0	..
White Bigarreau.....	85	0	..
<i>Prunus decumana</i> .....	59	0	..
Kentish Red.....	2,981	0	..
Reine Hortense.....	2,696	0	..

## PLUMS

## SELF-INCOMPATIBILITY

Many varieties of our domestic plums have been tested for self-sterility, and during the sixteen years that the work has been in progress many pollinations have been repeated. This has shown that degrees of self-fertility is a constant varietal character, and that the varieties dealt with may for convenience be placed in one or another of the following classes:

1. Those which are self-compatible and set a full crop with their own pollen.

2. Partially self-compatible varieties. In this class there is considerable variation, some set very few whilst others set a moderate crop when selfed, but all produce heavier yields when crossed with compatible varieties.

3. Self-incompatible varieties which wholly fail with their own pollen.

## CROSS-INCOMPATIBILITY

It may be recalled that as far as our work has advanced with cherries cross-incompatibility has always been reciprocal: e.g., if A fails with B, B always fails with A, and further, every other variety failing with A also fails with B. The incompatible cherries are also self-sterile, although as a rarity

an occasional fruit has formed. In plums two notable departures from the rule occur. 1. Incompatibility occurring in one combination whilst the reciprocal produces fruit freely. 2. Incompatibles which are partially self-fertile.

The individuals involved in this incompatibility and the groups to which they belong are:

## GROUP I

Coe's Golden Drop  
Coe's Violet  
Crimson Drop  
Jefferson  
Allgrove's Superb  
Seedling No. 1,024

## GROUP II

Seedling No. 1,026  
Seedling No. 1,030

## GROUP III

President  
Late Orange  
Cambridge Gage

## GROUP IV

Rivers Early Prolific  
Blue Rock

TABLE 2

SHOWING SELF- AND CROSS-POLLINATIONS BETWEEN INCOMPATIBLE PLUMS. THE NUMBERS AT THE TOP OF THE SQUARES ARE THE NUMBER OF FLOWERS POLLINATED AND THOSE AT THE BOTTOM THE NUMBER OF FRUITS WHICH REACHED MATURITY.

	Coe's Golden Drop	Coe's Violet	Crimson Drop	Jefferson	Allgrove's Superb	Seedling 1024	do 1026	do 1030	Late Orange	President	Cambridge Gage	Early Rivers Prolific	Blue Rock
Coe's Golden Drop	1226 0	200 0	366 0	1122 10	287 1	82 12	32 10	10 3	90 37	54 25	57 38	MANY MANY	35 17
Coe's Violet	73 0	733 0	141 1	585 1					195 88	44 31	71 49	163 MANY	
Crimson Drop	87 0	88 0	470 1	209 0									
Jefferson	868 1	414 2	515 1	352 0	220 0	90 42	29 23	13 10	16 15	53 21	15 11	MANY MANY	31 25
Allgrove's Superb	114 0	21 0		64 0	212 0	31 16	23 13		42 28		14 11	48 30	
Seedling 1024	71 0			274 0	103 0	240 0	32 10	8 5	60 14	54 18		50 18	18 9
do 1026	9 8			11 7		13 12	250 0	128 37	34 11	22 4	20 6	24 8	12 11
do 1030	29 13				17 7	19 8	100 1	90 0		12 3		24 10	18 5
Late Orange	97 36	94 32		78 19	30 6	21 5	27 10		604 0	549 0	194 54	78 46	29 22
President	116 60	74 38		75 28					559 0	509 0	209 61	18 11	116 12
Cambridge Gage		203 109							539 15	409 10	2625 42	15 10	
Early Rivers Prolific	45 11	226 86		57 23	83 27	65 24	56 9	21 8	246 102	91 39	126 57	8720 282	554 185
Blue Rock		126 96		36 23		25 16	12 2	7 7	103 66	49 39	63 41	388 17	1175 18

It will be seen in TABLE 2 that the four individuals printed in italics



above, i. e., Cambridge Gage, Blue Rock and Seedlings 1024 and 1030, fail on fertilization with pollen of the other sorts in the same respective groups, whereas in reciprocal fertilizations their pollen is effective. It is also noteworthy that in two of these, Cambridge Gage and Blue Rock, both the cross and self-incompatibility are only imperfect. Crimson Drop and Coe's Violet are well known to be bud sports from Golden Drop. More recently Allgrove's Superb has originated in a similar way from Jefferson and that it would behave similar to Jefferson in these tests was almost to be expected. That President fails when pollinated by Late Orange was originally reported by Rawes (1921). The variety Golden Esperen has in our experiments proved useless as a pollenizer. This, however, has only recently been used and is the only horticultural variety, whether in plums, cherries or apples in which such a high degree of male sterility has been evident.

A number of crosses have been made between varieties of *Prunus domestica* and *Prunus insititia*; all have resulted in full crops of fruit, indicating that they are completely inter-fertile. *P. domestica* pollinated with *P. spinosa* have with us wholly failed but from *P. insititia* crossed with *P. spinosa* we have obtained fruits from which seedlings have been raised. *P. domestica* when pollinated with *P. cerasifera* have occasionally formed fruits but the resultant seeds are usually imperfect, and from over seventy fruits obtained in this way only one has germinated.

The following list gives the classes of plums and the data obtained from the pollinations made.

## PLUMS THAT ARE SELF-INCOMPATIBLE

Variety	Number of Flowers Selfed	Fruit Set	% Set
Allgrove's Superb .....	212	0	..
Bryanstone Gage .....	594	0	..
Coe's Golden Drop .....	1,226	0	..
Coe's Violet .....	733	0	..
Crimson Drop .....	470	1	.2
Comte d'Althan .....	266	0	..
Descaisne .....	292	1	.3
Early Greengage .....	1,162	0	..
Golden Esperen .....	126	0	..
Jefferson .....	352	0	..
Kirke's Blue .....	1,133	0	..
Late Orange .....	604	0	..
Late Orleans .....	4,455	1	.02
McLaughlin's Gage .....	887	1	.1
Old Greengage .....	386	0	..
Pond's Seedling .....	932	0	..
Primate .....	36	0	..
President .....	509	0	..
Prune d'Agen .....	400	0	..
Frogmore Damson .....	1,536	2	.1
Transparent Gage .....	553	0	..
Yellow Magnum Bonum .....	429	1	.2

## PLUMS THAT ARE PARTIALLY SELF-COMPATIBLE

Variety	Number of Flowers Selfed	Fruit Set	% Set
Belgian Purple .....	822	62	7.5
Blue Rock.....	1,175	18	1.6
Cambridge Gage.....	2,625	42	1.6
Cox's Emperor.....	1,064	12	1.1
Early Orleans.....	1,798	25	1.4
Rivers Early Prolific.....	8,720	282	3.2
Utility .....	303	16	5.2
Farleigh Damson .....	809	60	7.4

## PLUMS THAT ARE SELF-COMPATIBLE

Variety	Number of Flowers Selfed	Fruit Set	% Set
Oullins Golden Gage.....	33	30	90.9
Denniston's Superb.....	253	163	64.4
Early Mirabelle .....	422	240	56.8
Myrobalan Red .....	76	43	56.6
White Magnum Bonum .....	191	100	52.3
King of the Damsons .....	246	110	44.7
Early Transparent .....	988	442	44.7
Pershore .....	32	13	40.6
Monarch .....	19	6	31.5
Prince of Wales.....	1,015	318	31.3
Victoria .....	3,916	1,185	30.2
Goliath .....	883	260	29.4
Guthrie's Late .....	1,541	367	23.8
Reine Claude de Bavay .....	26	6	23.0
Gisborne's .....	300	48	16.0
Belle de Louvain .....	487	63	12.9
Prune Geante .....	251	25	10.0

## APPLES

Preliminary cross-pollinations between certain varieties of apples suggested the view that compatibility—as far as the actual production of fruit is concerned—existed in a higher degree between certain varieties than others, but repetition and further work have disproved this view. For example, when we first pollinated Cox's Orange Pippin with Sturmer Pippin and Newton Wonder, but few fruits set, yet when in subsequent years we repeated these pollinations and also others which had behaved in a similar way, full crops of fruit were obtained. So far, we have not observed any clear example of complete cross-incompatibility in apples.

During the time these experiments have been in progress trees of many varieties have been successively used and in some varieties it has become evident that the degree of self-compatibility has increased with advancing age. Trees of several varieties which for a number of years had given wholly negative results have later formed occasional fruits with their own pollen. For example, in 1912 six trees of Cox's Orange Pippin of equal age and size were brought into the experiments; they flowered freely from 1914 onwards and large numbers of flowers have been self-pollinated in most seasons. In 1918 for the first time, two fruits set out of 816 flowers that were self-pollinated; since then these trees have always formed occasional fruits whenever they have been selfed. Prior to 1918 these and trees of other varieties which have behaved similarly had produced full crops of fruit when they had been crossed.

That varying degrees of self-compatibility exist in apples is evident, but to measure and to express this and also the results obtained from crossing is extremely difficult. This of course also applies to cherries and plums. The proportion of flowers which sets after compatible pollinations often varies as is abundantly shown in our records. Results have shown that the kind of crop the tree carried in the previous year and varietal differences in fruit size are frequently involved in these variations. A higher percentage of fruit is more often expected from a tree bearing a moderate number of flowers than from one which flowers freely. Indeed, when observing comparable examples it is not uncommon for the trees with the fewer flowers to mature the larger number of fruits; often this would be expected. These considerations show that intimate acquaintance, not only with varieties, but with the individual trees and the many factors involved, are essential to arrive at the real measure of fertility, and it is obvious even when many allowances are made that the actual percentage set can only be taken as an approximate guide. In experiments of this kind it is also evident that to obviate other courses of unfruitfulness the conditions under which the experiments are carried out, the health of the trees and attention to cultural details are of the greatest importance.

The majority of the varieties of apples that we have dealt with, although varying in degree, have proved to be only partially self-fertile. Among those so classified in the appended lists considerable variation occurs; some set very few fruits when selfed, whilst others set a moderate crop. Complete self-sterility has been of rare occurrence.

The varieties of apples classified as self-compatible have all eventually set full crops of fruit with their own pollen, but in their early years some of these only set a moderate crop when selfed, although they had set full crops when crossed. It therefore appears advisable in practice to provide for cross-pollination for all varieties of apples.

In many fruits arising from selfing certain varieties, e. g., Golden Spire, Lord Derby and Duchess of Oldenburg, there was no sign of a seed. Generally apples arising from self-pollination have contained fewer perfect seeds than fruits which have been crossed. Exceptions, however, have occurred in which crossed fruits have contained but few developed and viable seeds; which indicates that the combinations in which they occur are only partially inter-fertile.

In some varieties of apples, the shape of the fruit is strikingly influenced by the number of seeds developed, and occasionally fruits without seeds are smaller than those with seeds.

The following list gives the classes of apple varieties and the data obtained in the pollinations.

#### APPLES THAT ARE SELF-STERILE

Variety	Number of Flowers Selfed	Fruit Set	% Set
Brownlee's Russet .....	514	0	..
Gascoigne's Scarlet .....	47	0	..
Grimes Golden .....	36	0	..

## APPLES THAT ARE PARTIALLY SELF-FERTILE

Variety	Number of Flowers Selfed	Fruit Set	% Set
Royal Jubilee .....	1,017	1	.09
Winter Ribston .....	607	2	.32
Northern Greening .....	513	2	.38
Blue Pearmain .....	436	3	.67
Duke of Devonshire .....	229	2	.87
Lady Sudeley .....	197	2	1.0
Cox's Orange Pippin .....	5,101	53	1.03
Lane's Prince Albert .....	1,881	22	1.1
Newton Wonder .....	334	5	1.4
Beauty of Bath .....	1,769	30	1.6
King's Acre Pippin .....	370	6	1.6
Worcester Pearmain .....	858	16	1.8
King of the Pippins .....	522	10	1.9
Norfolk Beauty .....	210	4	1.9
Ribston Pippin .....	336	8	2.0
St. Edmund's Russet .....	935	19	2.0
Encore .....	132	3	2.2
Grenadier .....	200	5	2.5
Lord Derby .....	686	18	2.6
Bismarck .....	180	5	2.7
Annie Elizabeth .....	294	10	3.3
Lord Grosvenor .....	250	11	4.4

## APPLES THAT ARE SELF-FERTILE OR SELF-FRUITFUL

Variety	Number of Flowers Selfed	Fruit Set	% Set
Washington .....	1,187	61	5.1
Sturmer Pippin .....	135	7	5.1
Crimson Bramley .....	439	23	5.2
Golden Spire .....	989	56	5.6
Rev. W. Wilks .....	932	56	6.0
St. Everard .....	149	9	6.0
Red Winter Reinette .....	226	17	7.4
Antonowka .....	389	29	7.4
Bramley's Seedling .....	761	58	7.6
Stirling Castle .....	1,631	131	8.0
Cellini Pippin .....	160	14	8.7
Coronation .....	143	15	10.4

In the reports previously referred to (Sutton, 1918; Crane, 1923 and 1925) the results obtained from numerous compatible cross-pollinations in cherries, plums and apples have been published in detail.

## ECONOMIC ASPECTS

The practical conclusions to be drawn from the results of these investigations, and indeed from all such experiments, are self-evident.

Prospective planters will first consider the commercial properties of the varieties they select, but they should also find out which are self-fruitful; and if any varieties chosen are self-incompatible they should arrange for others which flower approximately at the same time and are known to be inter-compatible to be interplanted with them. There is no reason why varieties which are self-compatible to a high degree should not be planted in large blocks; but it is worthy of note that as far as we at present know the pollen of self-compatible kinds is always effective on self-sterile varieties, and therefore if any self-sterile kinds are grown the self-fertiles might be advantageously interplanted with them. In any case the planting of large areas of a



single variety, unless it is known to be self-compatible to a high degree, should be avoided, also the planting together of varieties which are cross-incompatible. Considered at the outset these precautions involve no additional expense and due attention to such essentials are likely to have a material bearing on future crops. Indeed, they may make all the difference between success and failure.

The appended lists show that practically all our consistently heavy cropping plums are self-fertile. This almost suggests that self-fruitfulness is one of the essentials of a commercial plum; that it is a valuable and desirable character is beyond dispute, but it has been clear throughout the course of these experiments that the majority of the self-sterile kinds have the capacity for high yields, e. g., the Coes, Jefferson, Pond's Seedling, President, Kirke's Blue and many others have repeatedly carried enormous crops when efficiently pollinated. Therefore, if such kinds were interplanted with suitable mates, and *an abundance of flower visiting insects assured*, there is no obvious reason why they should not be regular bearers and carry good crops.

Apart from damage due to frost and other causes, observations in the field commonly show that a large number of young fruits fall owing to lack of fertilization. The cause of such falls is doubtless attributable to the absence of flower visiting insects or to their enforced inactivity during bad weather. This demonstrates the potential value of judicious interplanting, and the enormous benefit likely to be derived from bees if they are maintained in orchards and thereby enabled to take full advantage of the bright intervals which frequently occur during a spell of inclement weather. *Fruit growers with extensive plantations should regard hive bees as an essential part of their equipment.*

#### CYTOLOGICAL AND GENETICAL OBSERVATIONS

The cytological investigations of my colleague, Mr. C. D. Darlington, (1926, 1927) may be summarized as follows:

The number of chromosomes (diploid) in *Prunus domestica* and *Prunus insititia* is 48. The number in *Prunus spinosa* is 32, and in "Marianna" believed to be derived from *Prunus Cerasifera* the number is 16.

Hybrids I have raised from *P. domestica*  $\times$  *P. cerasifera* and from *P. insititia*  $\times$  *P. spinosa* have the intermediate numbers expected, i. e., 32 and 40. It is of interest to recall that the pollinations we have made between *P. domestica* and *P. insititia* have always been completely inter-fertile, whereas from pollinations between *P. insititia* and *P. spinosa*, and especially from *P. domestica* and *P. cerasifera*, fruits with viable seeds are but rarely formed.

In cherries the orthoploid number of *Prunus avium* appears to be 16 and that of *Prunus cerasus* 32. The reduction divisions in the sour cherries, by their secondary pairing, show that *P. cerasus* is a true tetraploid, not derived simply from *P. avium* but differing from this species in possessing additional elements probably derived from *P. fruticosa*—also tetraploid.

Of about 30 varieties of domestic cherries examined nearly all by their aneuploidy, or lack of pairing, give evidence of their hybrid origin. It is therefore possible to consider our domestic cherries as resulting from recombinations of different series of eight homologous or partially homologous chromosomes—one in *avium* and two in *cerasus*.

The following shows the chromosome numbers (diploid) of the varieties and seedlings studied. Where irregularities in pairing of chromosomes have been observed in the pollen-mother-cell divisions the number of univalents and trivalents observed is indicated. It is evident that there is no direct association between the actual chromosome number and cross-incompatibility, e. g., Waterloo with three extra chromosomes is associated with Big. de Schrecken with one extra chromosome, both belonging to Group 2. Whereas Big. Napoleon and Emperor Francis with two extra chromosomes both belong to Group 3.

SOUR CHERRIES (32)		SWEET CHERRIES (17-19)	
Morello	2 univalents	Big Kentish	17 chromosomes 1 trivalent
Kentish Red	1 univalent	Big Noir de Schmidt	
Kentish Red "A"	and	Big Noir de Guben	
Wye Morello	1 trivalent	Big de Schrecken	
		Noble	
DUKES (32)		Elton	18 chromosomes 2 trivalents
Royal Duke	1 univalent and	Bohemian Black	
	1 trivalent	Emperor Francis	
Late Duke	2 univalents	Big Napoleon	
Empress Eugenie	2 univalents	Guigne d'Annonay	
Reine Hortense	6 univalents	Waterloo	3 trivalents
May Duke		Black Eagle	
		Knight's Early	
		Black	19 chromosomes

## SEEDLINGS

May Duke (32)  $\times$  Yellow Spanish (?16) = { Black Eagle (19)  
Waterloo (19) and Knight's Early  
Black (19)

Waterloo (19)  $\times$  Black Eagle (19) = seedlings with (19) and (16+) (tetrasomic)

Big. Napoleon (18)  $\times$  Big. de Schrecken (17) = seedlings with (18) and (16)

Big. de Schrecken (17)  $\times$  Black Tartarian "B" = seedling with (16)

Governor Wood  $\times$  Black Tartarian "B" = seedling with (16)

Kentish Red "A" (32) selfed = seedling with (32)

Wye Morello (32) selfed = seedling with (32)

Empress Eugenie (32) selfed = seedling with (32)

Morello (32)  $\times$  May Duke (32) = seedling with (32)

Big. Kentish (17)  $\times$  Morello (32) = seedlings with (32) and (24)

Wye Morello (32)  $\times$  Big. Napoleon (18) = seedlings with (23) and (24)

Elton (18)  $\times$  Wye Morello (32) = seedlings with (24) and (26)

Bohemian Black (18)  $\times$  Kentish Red "A" (32) = seedlings with (26)

It is noteworthy that all the sweet cherry varieties have extra chromosomes beyond the diploid number, and in view of the breeding results and known origin of Knight's seedlings we can have little doubt that hybridization with a tetraploid form has contributed to their origin. It is not possible to say whether certain members of the series alone have the extra chromosomes but the occurrence of tetrasomic forms in crosses between two of Knight's seedlings proves that these possessed one extra chromosome in common.

From the morphological point of view it seems to us that the influence of

*P. avium* is approximately equal to that of *P. cerasus* in the Dukes, and that the part played by *P. cerasus* in the constitution of the sweet cherries and by *P. avium* in that of the sour cherries must be considerable.

It is noteworthy that in selfed families we have raised from varieties of *P. cerasus* seedlings with *P. avium* characters frequently appear, and in families raised from crosses between varieties of *avium* occasional seedlings occur which show *cerasus* characters in a marked degree. Seedlings in families raised from *avium* crossed *cerasus* show a considerable variety of forms, only some of which resemble the Dukes in all their characters. These facts considered in conjunction with the cytological observations, and the common occurrence of aborted pollen and defective seeds in varieties usually regarded as pure *avium* strongly supports the above view. Moreover, they indicate that their bi-specific origin is fundamentally involved in the different forms of sterility that we have met with in cherries.

In plums our breeding results indicate that the properties of self-compatibility behave as a Mendelian dominant, although it seems probable that more than one genetic factor is involved.

TABLE 3

SHOWING SELF- AND CROSS-COMBINATIONS BETWEEN PARENTS AND SEEDLINGS. AT TOP OF SQUARES ARE THE NUMBER OF FLOWERS POLLINATED, AND AT THE BOTTOM THE NUMBER OF FRUITS WHICH SET AND MATURED.

		Jefferson	Allgrove's Superb	Comte D'Althan X Jefferson						Comte D'Althan
				1024	1026	1030	1025	1027		
Jefferson	{	352	220	90	29	13	12	14	45	
		0	0	42	23	10	8	11	22	
Allgrove's Superb	{	64	212	31	23				36	
		0	0	16	13				24	
Comte D'Althan	1024	274	103	240	32	8	45		72	
		0	0	0	10	5	19		37	
	1026	11		13	250	128		22	45	
		7		12	0	37		9	16	
	X 1030		17	19	100	90	15	20	30	
Jefferson	1025		31	8	1	0	4	8	9	
			153			19	107	14	41	
			31			4	1	7	16	
Comte D'Althan.	1027					66	31	610	74	
						19	10	4	6	
Comte D'Althan.	{	42		33	38	23	65	26	266	
		20		5	7	3	20	6	0	

It is noteworthy that all varieties involved in cross-incompatibility—both cherries and plums—are self-incompatible or nearly so. Self-fertile varieties pollinated *inter se* and self-steriles with self-fertiles have always proved compatible.

The behavior of the incompatible varieties and seedlings (see TABLES 1, 2 and 3) shows that self and cross-sterility in fruit trees is genetically complex, and that at the present stage of our investigations no simple interpretation is possible. Indeed, owing to the hexaploid constitution of our domestic plums, and the tetraploid and aneuploid constitution of cherries, self- and cross-sterility may well be more complex in these than in diploid plants.

Seedlings 1,024, 1,030, Cambridge Gage and Blue Rock, when used as females, fail on fertilization with pollen of the other sorts in the same respective groups, whereas in reciprocal fertilizations their pollen is effective. It appears probable that the individuals which are reciprocally incompatible and form a completely inter-sterile group are homozygous for the properties which determine their cross-incompatibility and that the individuals which give different results in reciprocal pollinations are heterozygous for these properties.

It also appears probable that such differences in reciprocal matings are due to the fact that on the female side of incompatible cross-pollinations we are not dealing with ordinary sexual crosses, but with a somatic phenomenon. The styles, where the pollen tubes are arrested, are of somatic tissue, consequently on the female side we are not concerned with the possibility of gametic segregation. In the pollen, however, we have gametic tissue and presumably plants heterozygous for incompatibility would produce grains of different kinds, some incompatible, and others compatible and effective in fertilization.

Histological investigations carried out in conjunction with Dr. E. J. Collins show that, in both self and cross-incompatible pollinations, the pollen tubes are arrested in the nutrient stylar tissue and fail to reach the ovary: consequently the young fruits fall at an early stage owing to lack of fertilization. Observations show that not only do the ends of the arrested pollen tubes themselves swell up, but in addition a slime sheath forms around them. We may therefore consider whether the arrestation is brought about by (a) an obstruction of a mechanical nature, or (b) an incapacity to absorb nutrient material owing to the presence of a layer of mucilage or allied substance forming a semi-permeable or impermeable membrane. Whatever the physiological nature of the arrestation may be, it is evident that self- and cross-sterility in plums and cherries is due to unfavorable reactions—probably of a very diverse origin—between the somatic tissue of the mother plant and the gametic tissue of the potential male parent.

The partial failures which occur at a later stage, and the formation of fruits with imperfect seeds are probably due to degrees of genetic incompatibility which arrests the embryonic growth at different stages. In the various manifestations of sterility in cherries we have to consider the probability



of a varying proportion of defective ovules and also the arrest of embryonic growth due to a lack of balance in the chromosome complement of the developing embryo.

Considering the numerous cherries which reach maturity without developing perfect seeds—in many practically only the empty seed coats remain—it is safe to conclude that if embryonic growth is not arrested until a fairly late stage the fruits, if favorable conditions prevail, are able to remain and reach maturity. There are, however, indications that at this stage the developing fruits are highly susceptible to any adverse influence, and are likely to fall in consequence.

### RASPBERRIES

In breeding experiments with raspberries, two horticultural varieties have when selfed given a proportion of plants which are completely impotent on their female side. These male forms occurred in selfed families raised from the varieties "Superlative" and "Norwich Wonder" as follows:

Superlative ♀		Norwich Wonder ♀	
Selfed		Selfed	
113 ♀	25 ♂	157 ♀	31 ♂

The hermaphrodite seedlings all have well developed female organs, but differences occur in the development of anthers and pollen. The large majority are functionally hermaphrodite, but a few individuals have very reduced anthers and so far have failed to produce any good pollen.

The male plants have obtuse, downfolded and undivided leaves on the two-year-old fruiting canes. In the young canes the leaves only have three lobes; they have comparatively short petioles, and are quite distinct from the divided leaves of hermaphrodite plants. This correlation between leaf characters and maleness enables the males to be classified at an early stage. Mr. Darlington has found that the diploid chromosome number of these different forms is uniformly 14.

The morphological characters of the leaves and the sex condition of these male plants approximates to *Rubus Idaeus obtusifolius* (Willd.), although according to Focke (1911, p. 208), *obtusifolius* forms occasional seeds from which weakly plants have been raised.

Both Superlative and Norwich Wonder have been reciprocally crossed with other hermaphrodite raspberries. From these crosses 309 seedlings have been raised, but no male forms have occurred.

### LITERATURE CITED

- Backhouse, W. O., 1911. Self-sterility and self-fertility in plums. Rep. Brit. Ass. 599.  
 Crane, M. B., 1923. Report on tests of self-sterility and cross-incompatibility in plums, cherries and apples at the John Innes Hort. Inst. II. Jour. Pom. and Hort. Sci. 3: 67-84.  
 ——— 1925. Self-sterility and cross-incompatibility in plums and cherries. Jour. Gen. 15: 301-322.  
 Darlington, C. D., 1926. On the cytology of the cherries. Rep. Brit. Ass. 1926: 407-408.

- 1927. The behavior of polyploids. *Nature* **119**: 390-391.
- Florin, R., 1923. Korsbarstradens Pollinering. *Sveriges Pomologiska Förenings Årsskrift* **1**: 1-31.
- Focke, W. O., 1911. *Species Ruborum*, *Bibl. Bot.* **72**: 208-209.
- Gardner, V. R., 1913. A preliminary report on the pollination of the sweet cherry. *Bull. No. 116, Oregon Agric. Coll. Exp. Sta.*
- Hooper, C. H., 1924. Notes on the pollination of cherries. *Jour. of Pom. and Hort. Sci.* **3**: 185-190.
- Knight, T. A., 1816. Some account of three new cherries. *Trans. Hort. Soc.* **2**: 208-211.
- 1818. Description and account of a new early black cherry. *Trans. Hort. Soc.* **3**: 211-213.
- Rawes, A. N., 1921. Self-fertility and self-sterility in plums. *Jour. Roy. Hort. Soc.* **46**: 353-356.
- Schuster, C. E., 1922. Pollination of the sweet cherry. *Oregon Agric. Exp. Sta. Cir.* **27**.
- 1925. Pollination and growing of the cherry. *Bull.* **212, Oregon Agric. Exp. Sta.**
- Sprenger, A. M., 1908. *De Onvruchtbaarheid der Kersenboomen in Zuidlimburg.*
- Sutton, I., 1918. Report on tests of self-sterility in plums, cherries and apples at the John Innes Hort. Inst. *Jour. Gen.* **7**: 281-300.
- Waite, M. B., 1894. Pollination of pear flowers. *U. S. A. Dept. Agric. Div. Path. Bull.* **5**.

### PLATE 8

A tree of Coe's Violet Plum about four weeks after the controlled pollinations were made. At *A*, pollination with Jefferson; at *B*, self-pollinated; at *C*, pollination with Bryanstone Gage. Note the large size of fruits with Bryanstone.

### PLATE 9

The same tree of Coe's Violet Plum shown in Plate 8 at a later stage, showing the final result. Fruit formed only on the flowers that were cross-pollinated with Bryanstone Gage. No fruit set to self-pollination nor to cross-pollination from Jefferson.

### PLATE 10

A tree of the Big Napoleon (Group 3) Cherry showing the result of incompatible and compatible pollinations.

At *A*, 258 flowers pollinated with Emperor Francis; no fruit set.

At *B*, 34 flowers self-pollinated, no fruit set.

At *C*, 154 fruits matured from 273 flowers pollinated as follows: 86 flowers × Guigne de Winkler, 42 fruits set; 37 flowers × Big. de Schrecken, 19 fruits set; 36 flowers × Roundell, 19 fruits set; 60 flowers × Belle de Orleans, 41 fruits set; 54 flowers × Early Purple Gean, 33 fruits set.

### PLATE 11

A tree of the Early Rivers (Group 1) Cherry used in the experimental study. No fruits developed to self-pollination nor to cross-pollination with Black Eagle, Bedford Prolific, or Knight's Early Black. Cross-pollinations with five other varieties set fruit as indicated.

### PLATE 12

A tree of the Royal Jubilee Apple. At the left side and the top of this tree 151 flowers were self-pollinated; no fruit set. At the right hand side 62 flowers were crossed with Lane's Prince Albert and of these 10 fruit set.



CRANE: STUDIES IN STERILITY

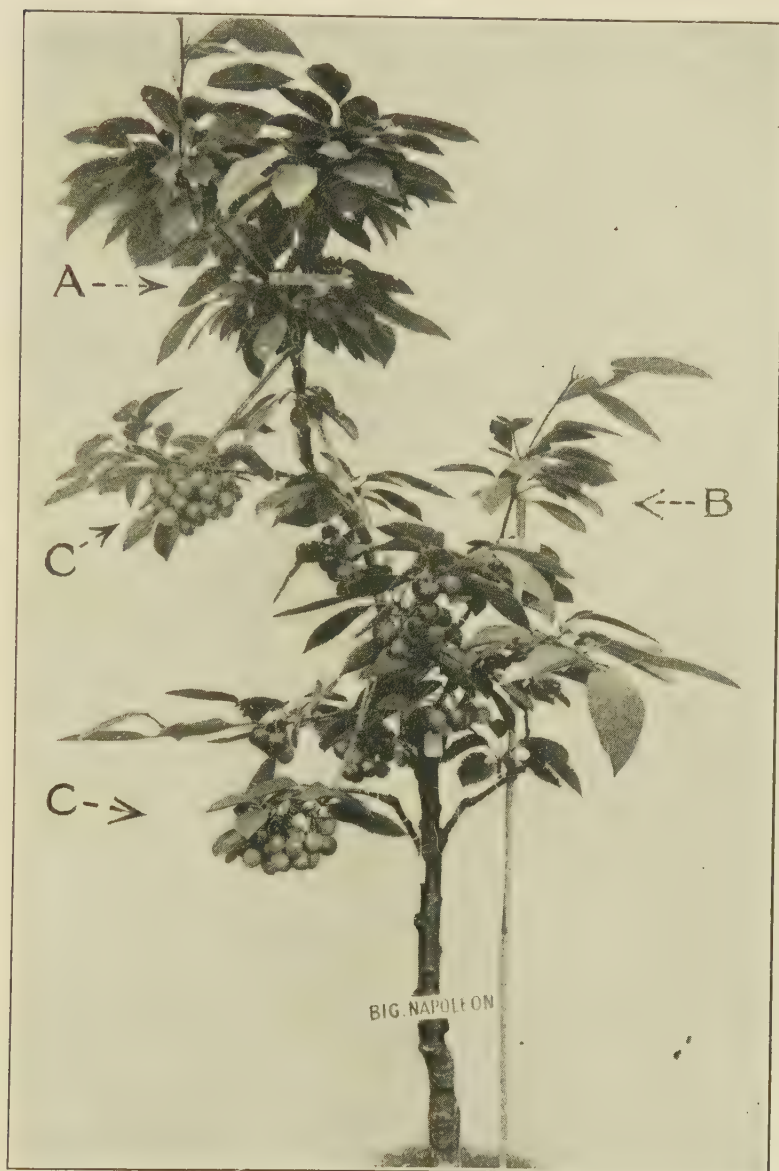






CRANE: STUDIES IN STERILITY





CRANE: STUDIES IN STERILITY







CRANE: STUDIES IN STERILITY





CRANE: STUDIES IN STERILITY





# SOME FACTORS TO BE CONSIDERED IN THE PRACTICAL APPLICATION OF STERILITY STUDIES OF FRUITS

A. J. HEINICKE

*Cornell University*

In comparison with the striking results frequently derived from cross-pollination in orchards, the many other factors concerned in the set of fruits appear relatively unimportant. It may seem impertinent therefore to call attention to the need of a more reasonable basis for evaluating the various conditions that influence the set of fruit. But if we neglect the study of the specific influences of each of the numerous factors, how can we be certain that the small crop often obtained with self-pollination could not be increased by modifying some of the other conditions as well as by supplying foreign pollen? Unless such investigations go hand in hand with the sterility study of orchard fruits, there is danger that much of our data regarding the need for cross-pollination and the information concerning compatible and incompatible varieties may be misleading in orchard practice.

Some work has been done by the Department of Pomology at Cornell regarding the influence of nutrition and water supply on the set of fruit, and a few of the results have been published from time to time (2, 3). For the purpose of this paper it may be sufficient to cite several specific cases in which the importance of some of these factors of nutrition and water supply seem to be paramount.

The Anjou pear is known to be an uncertain cropper and is generally listed as self-sterile. But it frequently fails to set fruit even in orchards having other varieties nearby. A typical case was that of Mr. Tupper's orchard in Tompkins County, N. Y. His twenty-year-old Anjou orchard adjoining a block of Seckel pears had never produced satisfactory crops and the owner was about to cut down the trees. In the spring of 1923 some trees were pruned moderately; some so heavily that the bearing surface was practically eliminated; some of each treatment received five pounds of sodium nitrate to the tree. Suitable checks without treatment were available. The entire orchard was cultivated and sprayed.

In 1923 the crop, though small, was the best ever obtained. There was much more fruit in 1924, and a very heavy crop in 1925. The bulk of the crop in all years was secured from the heavily pruned trees, though nitrogen with light pruning gave some increase over the checks. The unpruned trees, even when nitrogen was added, continued to bloom without setting much fruit. This year, 1926, both the Seckels and the Anjous have a light

bloom, and, as a result, there is a small crop. In this case then the effects of the pruning lasted for at least 3 years.

Practical growers have learned that Anjou pears need very much heavier pruning than most other varieties, although some are still skeptical about the need for cross-pollination. In several cases the heavy pruning incident to top-working the Anjou to a different variety has caused the remaining branches to bear satisfactory crops, even before the variety intended to supply foreign pollen produced blossoms. Unless one takes into consideration the special cultural needs of a variety, as for example, heavy pruning in this case, it is easy to understand how the variety may be classified as self-sterile or self-fertile, or even as incompatible with other varieties.

The second case is that of Mr. Dickerson's orchard in Niagara County. Rhode Island Greening and Wealthy apples about 15 years old are interplanted in this orchard, the trees being 20 feet apart. The Wealthy apples have good crops whenever they bloom, but the Greenings produce very few apples even though they bloomed heavily each year. They have been well cared for and made excellent terminal growth and appeared to be healthy and normal when examined this spring and during the summer. Adjoining Greening trees with Baldwin interplanted have heavy crops whenever they bloom.

The problem of unfruitfulness of the young Greening tree appeared to be one of inter-sterility of the two varieties, Wealthy and Greening. Accordingly this past spring, branches with blossoms of other varieties—Ben Davis and Baldwin—were placed in buckets of water about the orchard, and hives of bees were placed nearby to insure distribution of the pollen. The bloom was very good and uniformly distributed.

Even though there was only one day of good pollination weather during the blooming season, the crops are again heavy on the mature Greenings, and the heaviest they have ever been on the young Greenings interplanted with Wealthy. But the crop is no better on the young Greening trees which had the benefits of introduced pollen than it is on the rest. Furthermore the crop is very scattering, and found mostly on certain of the lower limbs.

This latter fact seemed especially interesting. Closer examination revealed that in practically every case the heavy fruiting limbs which were 3 to 4 inches in diameter at the point of origin had several good-sized branches (1 to 2 inches) removed near the trunk during the dormant season of 1924-25, that is a year ago the past spring. No doubt there were some pruned branches which did not stand out on account of their crop, but in all cases observed the heavy set could be attributed to the influences of the pruning.

Mr. Dickerson also has a solid block of twenty-year-old Greenings planted 20 by 20. These have been heavily fertilized and made a satisfactory growth in previous years, but this year, without fertilization, they made much less growth and appeared undernourished. The soil seemed none too strong and was undoubtedly exhausted by the peach fillers that had been removed several years ago. The owner planned to remove every other diagonal row of

Greening trees in the fall of 1926 and in preparation he cut off many of the large low branches in the spring of 1925. These pruned trees in the solid block of Greenings have a fair crop as have the outside rows along the lane. The trees not pruned this spring, though pruned in the past, and by no means presenting a neglected appearance, have very few fruits.

The influence of pruning in increasing the set of fruits on trees that bloom has also been noted in young McIntosh trees at the Cornell Orchard at Ithaca, and in a 20-year-old Northern Spy orchard in Orleans County. Unusually severe pruning such as accidentally occurs when a main limb is broken from the tree, frequently causes the remaining portion to set a heavy crop of fruit. Such cases afford excellent opportunity to study the changes in water supply and nutrition and may frequently give us important leads to possible modifications in our orchard practice. The beneficial effect of pruning on the set of fruit is not to be confused with the delay in bearing caused by the same practice on healthy young trees that have not yet reached the age at which they begin to bloom and which are still in need of a good bearing surface with well-developed spurs.

The third observation will serve to emphasize the influence of the preceding crop on the one following. The crop of McIntosh in New York State promises to be much smaller in 1926 than usual. Many McIntosh trees that fruited last year did not bloom this spring, but in at least one important fruit section, in the Champlain Valley, the trees bloomed heavily. They failed, however, to set satisfactory crops. In the extensive orchard of the Northern Orchard Company at Peru, N. Y., there are large blocks of the variety McIntosh and one might suspect that the lack of cross-pollination would be a factor in that case. But the orchard has done well in the past, and has received very good care. Furthermore, the crop seems no better where the McIntosh rows are adjacent to the variety Tolman Sweet, nor in the vicinity of several odd trees scattered through the orchard. The weather at blooming time, though not ideal, could not be held responsible since occasional trees in the orchard had good crops, and it was generally reported that the older McIntosh trees throughout the section were bearing satisfactorily. This fact seemed of special interest and inquiry showed that most of these old trees had very poor crops last year, 1925. In at least a few cases it was definitely recalled that the young trees with a good crop in 1926 likewise had few apples last year. It is not unreasonable to suspect, therefore, that the occasional fruitful trees scattered throughout the young orchards were probably deriving the benefits of an off-season in 1925.

Even though last year's crop did not interfere with the flower bud formation in the Champlain section as it did in other parts of the state, it evidently affected the nutrition of the tree and the development of its various tissues in such a way that the flowers could not remain on the trees in normal numbers.

Another case may be cited which throws light on a rather difficult problem. Several years ago one of the Cornell students took charge of an orchard

in Nassau, Del., in which there was a block of Stayman Winesap trees. Dr. Auchter has conclusively demonstrated that under Maryland conditions the set of fruit will be greatly benefited by cross-pollination with suitable varieties (1). But I am told that the Stayman orchard at Nassau did not give satisfactory crops even with suitable pollinators. Finally half of the trees were removed from the Stayman block, after which the variety fruited heavily throughout the orchard. I have not seen this orchard myself, and am not familiar with all the circumstances. I am told by the extension pomologist, Professor Snyder of Pennsylvania, that he had observed similar striking effects in solid blocks of Stayman from which half of the trees were removed. In Mr. Dickerson's orchard in which the Wealthy and Greening trees were interplanted, some of the Wealthy trees had died several years ago. This year the surrounding Greenings had good crops on the side nearest the missing trees. It seems as though the greater feeding area, the lack of competition and better exposure may help the set of fruit of varieties inclined to be self-sterile.

These few cases will serve to emphasize that all factors that have a direct or an indirect influence on the supply of water and other growth producing substances for flowers and fruits, including conditions such as weather, soil treatment, spraying, pruning, pollination, seed formation and the like are important for the final set. Although pollination, and frequently cross-pollination, may be regarded as an essential step in the formation of fruits, it must be remembered that effective pollination is often impossible because of unfavorable weather or other conditions that interfere with the work of pollen-carrying insects. Since we cannot depend too much upon cross-pollination as a means of insuring a good crop, it therefore seems wise to give more attention to the factors that are favorable for a set of fruit even though the weather is not ideal.

#### LITERATURE CITED

1. Auchter, E. C., 1921. Apple pollen and pollination studies in Maryland. *Pro. Amer. Soc. Hort. Sci.* 1921: 51-80.
2. Heinicke, Arthur J., 1917. Factors influencing the abscission of flowers and partially developed fruits of the apple. *Cornell Agr. Exp. Sta. Bull.* 393.
3. ——— 1919. Concerning the shedding of flowers and fruits in apple. *Proc. Amer. Soc. Hort. Sci.* 1919: 76-83.



# AN EVALUATION OF CERTAIN METHODS USED IN THE STUDY OF THE POLLINATION RE- QUIREMENTS OF ORCHARD FRUITS

L. H. MacDANIELS

*Cornell University*

In a field of research as broad as that of plant sterility, it is not only natural but inevitable that many diverse points of view are represented among the workers in that field. On the one hand, there are the more technical phases of the problem which hold the attention of the cytologist and the geneticist, and on the other, the more practical aspects which concern the fruit grower directly in the production of profitable crops. In the very considerable amount of research that has been done in an effort to solve important practical problems in different fruit sections of the country real progress has been made. It is to be expected, however, that with work done under such diverse conditions many of the results are scarcely comparable because of differences in the methods used or in some cases of the failure of workers to state the conditions of the experiment which are needed for the intelligent interpretation of the data presented.

The purpose of the present paper is not to destructively criticize the work or methods of any particular worker. Neither is there any intention of attempting to standardize the methods to be employed in future pollination studies as such standardization is neither practicable nor desirable. It is rather to point out the weaknesses in some of the methods in use, to call attention to factors which should be considered in connection with all methods, and to emphasize the importance of stating the conditions of an experiment in a sufficiently accurate way to aid in the interpretation of data. It is of course realized that most, if not all, of the points brought up have been considered by many workers though sometimes it has not been made clear that such was the case. It is also borne in mind that Chandler (3) and others have ably criticized some of the methods employed in pollination work. Nevertheless it is thought, in view of the fact that there are methods in use that have not been reviewed, a paper of this sort may serve a useful purpose if it does no more than direct the attention of workers to a more critical attitude toward the methods employed.

As in the case of practically all characters of living organisms, self-fruitfulness cannot be considered as a constant characteristic of any variety. It may vary within rather wide limits depending upon such a host of factors, any one of which may be limiting, that it is often impossible to ascertain which may be responsible for any given behavior. The ability of a variety to set fruit with its own pollen or with the pollen of another variety ap-

parently may vary from almost total self-sterility or inter-sterility, on the one hand, to a satisfactory degree of self-fruitfulness or inter-fruitfulness on the other, depending on environmental conditions. And different varieties show different degrees of self-fruitfulness. Some, like the Rhode Island Greening, the Stayman and the Winesap, have been consistently self-barren in so far as the available data show, whereas others like the Baldwin seem to have a rather high degree of self-fruitfulness under some conditions, but not in others. It is quite probable that under very favorable conditions and with improved experimental technique many of the varieties that seem to be self-barren on the basis of our present data would be found to set fruit.

### EXPERIMENTS WITH THE BALDWIN

The results of some pollination experiments with the Baldwin apple in different parts of the country give an interesting example of the different behavior of a variety under different conditions and with different methods. As the variety is grown under cultivation in Western New York, it apparently sets good commercial crops whenever it blossoms, even under weather conditions that in many cases are unfavorable for cross-pollination. The variety is frequently planted in solid blocks of considerable size and yet there is rarely, if ever, crop failure that seems attributable to lack of pollination. Rhode Island Greening, on the other hand, is self-barren to a greater degree and, though set in no larger blocks than Baldwin, frequently fails to set commercial crops when the weather at blooming time is unfavorable. In pollination experiments at Ithaca, New York, in 1925 (10), two dwarf Baldwin trees caged with bees gave essentially the same set as open pollinated trees. With all of these trees, however, the bloom was light and this was probably a factor in causing so large a set.

A. L. Pierstorff (unpublished), working in Western New York to find the effect of pollination upon the susceptibility of flowers to infection by fire blight, has incidentally obtained some interesting data on the self-fruitfulness of the Baldwin. His method was to cover branches before the blossoms opened with cheesecloth bags three feet long. When the blossoms were open they were selfed by brushing the stigmas with a camels'-hair brush, holding pollen from the same flowers. No attempt was made to select spurs. The bags were of course replaced immediately. The count of the fruits set was taken just before the June drop. In no one of the four years did he fail to get a good set of fruit. In 1926 in six bags containing 452 blossoms he obtained 85 fruits or 18.8 per cent set. This percentage is lower than it normally would be because in each case there were some blighted blossoms which were killed. The bloom was fairly heavy in all cases.

Overholser (12), in work reported at this conference, obtained a set on selfed Baldwin blossoms of 7.1 per cent, which, provided the bloom was fairly heavy, would be a fair commercial set. Sutton, in England, states that the variety set well when selfed, but whether or not a commercial set was obtained is uncertain. The work of Gowen (5) and Lewis and Vincent (9) with the

variety Baldwin is difficult to interpret. Indications are that the set obtained would not be considered adequate commercially.

Howlett (unpublished), who has made a very extensive and careful study of the Baldwin variety in Ohio over a period of several years, finds that variety unable to set a satisfactory commercial crop either on flowers which are selfed and bagged or upon a tree which was caged with bees, and Auchter, in Maryland, Morris (11) in Oregon, Sax (13) in Maine, and Waugh in Vermont obtained a set on selfed blossoms so small as to be negligible from a commercial standpoint. However, the number of blossoms covered by Gowen and Waugh was hardly enough to be significant and in the work of Auchter, Morris and Waugh the blossoms were bagged but not pollinated, a practice which in some cases at least does not assure self-pollination.

Where Baldwin pollen has been used on other varieties there are also contradictory results. Thus in Western New York, where Rhode Island Greening and Baldwin are frequently planted together, Greening rarely fails to set a crop, provided weather conditions are favorable for pollination at blooming time, even though Greening is recognized to be practically self-barren. In the spring of 1926 in Western New York, in some cooperative experiments carried on by the Cornell Pomology Department, branches of Baldwin blossoms in pails of water were placed in trees in a large block of McIntosh which had not fruited well, although the trees had blossomed heavily. A number of other varieties were placed in other parts of the orchard. A marked increase in the set of fruit was observed near the blossoms of all varieties tried except Tompkins King, even though the weather had not been particularly favorable for pollination, and Baldwin was apparently as effective a pollenizer as any variety used.

In the orchards at Ithaca in 1926, Baldwin pollen failed to give a good commercial set on caged McIntosh and Greening trees and yet in the same season in the same orchard in work done by R. Snyder (unpublished) when Baldwin pollen was placed on a single flower on each of many vigorous spurs in competition with flowers pollinated with Delicious Greening and Cortland on the same spur, Baldwin gave as good a set as Delicious and better than the other two varieties. About 70 per cent of the spurs set fruit with some one of the four varieties and 24 per cent set with Baldwin. On spurs holding more than one fruit Baldwin occurred more times than any other variety.

Howlett, working in Ohio, has used Baldwin pollen on many varieties over a period of several years with uniformly poor results and Overholser (12) found Baldwin pollen to give no set on Gravenstein in California. Gowen (5) and Sax (13) also find the variety a poor pollenizer as have others.

#### FACTORS AFFECTING FRUIT SET

The underlying cause for such differences in results are difficult to determine. Manifestly they are attributable to either differences in environmental factors including conditions of vigor and growth, faulty technique, or differ-



ences in the methods of experimentation which affect the setting of fruit. It is highly probable that environmental factors are by far the most important. As Heinicke (7) and others have pointed out, adequate pollination is only one of the many factors concerned in determining the set of fruit and in case of any failure to set may or may not be the limiting factor. There are abundant instances of apparent self- or inter-sterility in which it has been shown that failure to set after full blossoming has been due to causes other than inadequate pollination.

In carrying on pollination experiments in a practical way it is naturally of the utmost importance that all factors that may be limiting to fruit setting be eliminated in so far as possible. Some of these factors such as temperature, prolonged cloudiness, rainfall, sunshine and the like cannot be controlled, but should of course be accurately observed and the conditions reported with the data, especially if their effect was likely to be adverse to fruit setting. Another type of environmental factors of importance are those that might be more or less definitely controlled, but in the present state of our knowledge cannot be accurately evaluated. Here belong such practices as pruning, the application of fertilizers, especially nitrogen, and cultural practices, e. g., growing the trees under cultivation or sod. These controllable conditions should all be made as favorable for fruit setting as possible so that no one of them would be limiting as to fruit setting and the results obtained would be attributable to differences in pollination alone. We do not know enough about the effect of these practices on fruit setting to use them in regulating fruitfulness with any confidence that we are doing the most effective thing. Of course, diseases and insects, for the most part controllable factors, may be important for the direct damage they do.

Certain conditions in the trees themselves which affect the setting of fruit cannot be controlled directly, but by choosing for experimentation trees in a similar condition in so far as the experimenter can judge, sources of error can be avoided in part. Thus the pollination results on a tree with a heavy bloom should not be compared with those with a light bloom for a higher per cent set would be expected on the tree with a light bloom. Also one should consider whether or not the trees used in an experiment bore heavily the preceding year for if so the set is likely to be light, at least with some varieties. McIntosh trees in New York have been observed to fail to set crops even with a heavy bloom if the trees bore the year preceding, whereas trees that did not bear heavily the preceding year set good crops under conditions that were otherwise the same. The choice of vigorous spurs is always advisable, especially in hand pollination work, for time and labor spent on weak spurs is likely to be wasted because no flowers would set on them even under most favorable conditions. It would seem superfluous to say that any tree showing signs of winter injury or other weakened condition should be discarded but it should be borne in mind that such weakened condition is not always apparent at blooming time and that great care is necessary in choosing trees or limbs of trees upon which to work.



Little can be said regarding poor technique, although it is probably a factor in some cases. Some workers are good technicians and others are poor, and in a method that involves so delicate an operation as the emasculation of blossoms, this will always be a source of discrepancy in results. Contamination of the pollen supply is always a possibility, but with careful workers is undoubtedly safeguarded so that it rarely occurs.

### EXPERIMENTAL METHODS

An ideal method for studying pollination problems in a practical way under reasonably well controlled conditions is yet to be found. Such a method would not change the normal physiology of the tree, blossoms or foliage in any way but would allow perfectly natural development of flowers and fruit. It would make possible the use of large numbers of flowers on a considerable number of trees. It would provide checks which were sufficiently numerous and which did not differ in nutritional relationships from the blossoms pollinated. It would not involve a difficult or exacting technique.

All of the methods now in use are unsatisfactory from some standpoint though some are less objectionable than others. The method most used in pollination studies is that of the emasculation of the flowers by hand and the pollination of the stigmas with a soft brush laden with pollen usually covering the blossoms with opaque or "glassine" paper bags for a longer or shorter period while the stigmas are receptive. Of course, in determining the self-fruitfulness of a variety emasculation is not necessary though, according to the best evidence hand pollination is advisable in such cases. Usually the petals, stamens and outer part of the calyx tube are removed together, with the nails of the thumb and finger.

Several variations or refinements of this method are in use. Overholser (12) found that pollinating the stigmas by touching them with the bottom of the cork of the pollen vial was very satisfactory because it was possible to see where the pollen had been removed from the surface of the cork by the stigmas. Howlett (unpublished) reduced the number of blossoms to two laterals on vigorous spurs, emasculated these with a sharp scalpel and applied pollen by placing the small pollen-containing capsule over the stigma and giving it a slight twist. Such choice of spurs and thinning of blossoms would be expected to give a very large percentage set as compared with methods in which all flowers on the spur were used.

Snyder (unpublished), working in the Cornell orchard in 1926, chose very vigorous spurs of McIntosh, thinned the blossoms to four laterals, emasculated these and to each of the four blossoms of the spur, applied a different variety of pollen at the same time. This method was used in an effort to find out which of the four kinds of pollen was most effective in pollenizing McIntosh. Each flower on the spur was labelled with the name of the pollen variety by slipping a string label around the pedicel. The fact that the four different flowers were as nearly alike as possible in that they were on the same spur and that they must compete directly with each other for water

nutrients would seem to eliminate many of the variable factors found in other methods.

There is no question but that much valuable data have been obtained by the hand pollination method and where the results have not been negative many of the conclusions drawn regarding the self-fruitfulness or inter-fruitfulness of certain varieties have probably been justified, especially if the results have been consistent over several years and involved a considerable number of trees. On the whole, it is probably the best controlled method now in use. Yet the method is open to serious criticism on a number of points in that many sources of experimental error are introduced. One of the most serious sources of error is the operation of emasculation itself. There is no question but that with a skillful operator little or no damage is done to the tissue of the flower. Probably the operation is done with the least damage when a thin sharp scalpel is used though it is often done very successfully with the finger nails. Results obtained with different varieties by a single worker would of course be consistent as the error introduced would be the same, but where the work of different investigators is compared the difference in technique may be an important factor in causing differences in results. It would be an interesting study to obtain further data on this point by comparing the results of a group of graduate students working under the same conditions.

Probably in no other method is the personal equation of greater importance not only in the matter of technique, but because of the great chance for variation in the choice of the type of spurs selected for pollination. A single worker using his best judgment cannot select spurs that are equal in their ability to set fruit. If he could, he might by careful selection get all of the spurs to set. With persons whose conceptions of vigorous spurs are not alike in the first place and with trees that are unlike in their nutrition, it is practically inevitable that results will not be comparable.

Another difficulty with the method is that it is slow, making it hard for any one worker to use a very large number of flowers. This is one of the chief criticisms of much of the earlier hand pollination work. Some of the recent workers, however, by the use of great diligence or considerable help, have used many thousands of blossoms and obtained valuable results.

The shading of the spur leaves by covering the blossoms with bags has been referred to by Ewert (4), Heinicke (6), and others as a possible source of error. The possible effect of the bag is of course greatly reduced by leaving it over the spur for the shortest possible time and using transparent paper such as "glassine." Where consistently good sets have been obtained with certain varieties under bags it is not probable that the bags have made any significant difference. However, in cases where there is a very delicate balance of factors determining whether or not the blossoms on any given spur will set or fall, the effect of the shading may be very important. Certainly shading the leaves on a spur must have a very profound influence upon its physiology, at the very time that is most critical for the setting of the

blossoms. At least photosynthesis would be greatly reduced, which must have some influence on the nutrition of the flowers. In addition the humidity and temperature would also be increased within the bag. The temperature effect would probably be most noticeable in opaque bags. Heinicke (unpublished) finds that shading the spur leaves with a bag even for a day increases the catalase activity of the tissue in a striking manner. Although the significance of catalase activity is not altogether understood there is a high correlation between high catalase and vigorous active tissue and in most cases a change in the rate of catalase activity indicates a change in metabolism. It would seem that we know too little about the possible effect of shading by bags to rule it out as a negligible factor.

In order to obviate the possible effect of shading, certain workers have not covered the blossoms after emasculation because, with the petals removed, Lewis and Vincent (9) have shown that no insects were attracted to the flowers. Sax (13), in Maine, found this method fairly satisfactory though on one McIntosh tree near other varieties some cross pollination was indicated. Howlett (unpublished), working in Ohio, also obtained good results without bagging, the per cent being about the same with emasculated flowers inside and outside the bag. Overholser (12), however, working in California, found that without the protection of the bag the emasculated flowers dried out through the exposed broken tissue. This indicates at least that this method is not generally applicable and under some conditions would not reduce the experimental error, though in other cases it might do so. It is the writer's present opinion that practical problems of orchard pollination can never be satisfactorily settled by the emasculation, hand-pollination, bagging method although it is of great value in indicating relationships which can be further checked up with more extensive field trials under more natural conditions.

Another method which has been used by a number of workers, Hendrickson (8), Alderman (1), and others, is to enclose whole trees in frames covered with light cloth or mosquito bar to exclude insects and to place a hive of bees within to insure pollination. There are obviously a number of real advantages in this method. Among the most outstanding is the ability to use very large numbers of flowers with much less skilled labor than in the case of hand pollination work. The frame work of the cages can be put up before the blossoms open, thus obviating the mad rush during the blooming period. It would seem also that blossoms so treated must be under more natural conditions than those emasculated and bagged, the shading resulting from mosquito bar, though appreciable, not being nearly so dense as that of the bags usually employed and the mutilation of the blossoms by emasculation being prevented.

On the other hand, there are distinct disadvantages in the method. In the first place, because of the expense of the cages, only a small number of trees can be enclosed, usually only one of a variety, and results based on experience with a single tree are always open to criticism. Secondly, if a strong colony of bees is placed in the cage the blossoms receive far more visits than



would be possible under orchard conditions and we do not know what the effect of this may be. It is quite possible that under such conditions the set of fruit would be far above what might be expected under normal conditions. Howlett (unpublished) finds this method gives a 3 to 5 per cent better set with the Baldwin than hand pollination. On the other hand, it is conceivable that if the weather is favorable for pollination the blossoms may be visited so often that injury to the stigmas may result from the rough treatment given by the bees.

In work done at Ithaca in 1925 (10) and 1926 consistent results have been obtained with the cage method in cases where it was possible to place abundant pollen of a variety into the cage during the early part of the blooming period. However, if the bees had a chance to work upon the blossoms of the caged tree before the introduced pollen was shed the stigmas of the caged tree were apparently rendered unreceptive due possibly to mechanical injury but more probably to the effect of self-pollination on a variety that was practically self-sterile. Thus in 1925 a McIntosh tree was caged with bees which were allowed to work selfing the blossoms until the tree was nearly in full bloom for the purpose of pollinating the check limbs. These were then covered with large bags and Greening pollen was introduced. The set of fruit on this tree was no greater than on an adjacent caged McIntosh tree which was selfed. In 1926 a McIntosh tree was caged with bees and R. I. Greening pollen put in at the beginning of blooming. This tree set a good crop with the Greening pollen. In 1926 also a McIntosh tree was caged with bees and Baldwin blossoms introduced. These failed to shed pollen until after the McIntosh blossoms had been selfed by the bees. A very small set resulted although it is certain that the stigmas of covered blossoms on the same tree had not ceased to be receptive during the time that the Baldwin pollen was available. This would argue for the inter-sterility of Baldwin and McIntosh but for the fact that Baldwin pollen gave a good set on McIntosh in hand pollination experiments indicating that an explanation other than inter-sterility was probable. We need to know more about the effect of excessive work of insects on the stigmas, about the possible effect of shading by even light cheesecloth or mosquito netting, about the length of time during which stigmas are receptive and the factors influencing such receptivity before the results of this method can be satisfactorily interpreted.

Another method which has some good points is to cover whole limbs of 1 to 2 inches in diameter with cheesecloth bags a yard wide and 5 or 6 feet long. A number of bags can be put upon the same tree and any number of trees can be used. The bags can be removed and the blossoms selfed or crossed with a brush early in the morning or late in the evening when insects are not working or at any time, if care is taken to keep insects away from the blossoms when they are exposed. An especially good feature of this method is that it is possible to choose, as checks, limbs coming from the same section of the tree or even from the same main branch. Large numbers of blossoms can be used without emasculation. From the standpoint of practical orchard



pollination problems this method is fairly adequate in that it is possible to determine whether or not a variety is self-fruitful or not. If the variety is self-fruitful to a sufficient extent to set commercial crops with its free pollen that is all the orchardist wants to know. If shown to be self-unfruitful, the variety can be pollinated with pollen from other varieties without the troublesome operation of emasculation.

One drawback to the method is that there is still the largely unknown effect of shading to deal with, although by putting on the bags immediately before blossoming and removing them at once after the stigmas cease to be receptive this is reduced to a minimum. Also there is the danger of insects getting access to the branch while the bags are temporarily removed for pollination. The same possibility of contamination of the pollen supply exists as with any hand pollination method.

Whenever varieties suspected of being self-sterile are planted in large blocks one of the most satisfactory ways of testing the value of cross-pollination or of inter-sterility relationships is to introduce pollen of other varieties in different parts of the block by placing branches an inch or more in thickness of the pollen variety in pails or cans of water and suspending the cans in trees that are blossoming well. A supply of bees must be provided if insects are thought to be lacking in the locality. If the variety introduced is a good pollenizer for the orchard variety a very marked increase in the set will be observed near the introduced blossoms. This method has been used very effectively by Auchter (2) in Maryland and in 1926 by members of the Pomology Department of Cornell. It is of great value in checking up the results of other methods as in this case the conditions are more nearly normal than with any other method.

Obtaining a markedly better set near the introduced blossoms than on adjoining trees is satisfactory evidence that the variety is a good pollenizer under orchard conditions, provided the set is large enough to be adequate commercially. Negative results as always are open to question.

### PRESENTATION OF DATA

A considerable part of the confusion in the evidence on pollination experiments has been due to the incompleteness of the data given. One of the most misleading expressions is that of percentage set referring to the number of fruits obtained in a given experiment divided by the number of blossoms pollinated or counted. Taken by itself this means absolutely nothing as to whether or not a satisfactory commercial crop was produced and may mean very little as to the possible inter-sterility relationships of the varieties involved. In order to be significant it is necessary to know what percentage of the spurs or growing points on the tree blossomed, how many spurs set fruit and in case of hand pollination experiments, to what extent vigorous spurs were selected. A 10 per cent set on a tree with 15 per cent of the spurs blossoming is a far different matter than the same percentage setting with 70 per cent of the spurs blossoming. In the former case the number of apples

borne is insufficient to give a commercial crop and the percentage set is probably considerable higher than it would have been had there been a heavy bloom. Obviously also a set on 10 per cent of spurs selected for their vigor gives very little information concerning a possible commercial set or concerning the value of the variety as a pollenizer under field conditions where all blossoms are to be considered. It is also of great importance that the data regarding set be taken after the June drop and preferably at maturity because with many weakly self- or cross-compatible varieties, fruits may hang on through the June drop and then fall because of lack of seeds or poorly developed seeds resulting from unsatisfactory pollination. Gowen (5) has pointed this out for Maine conditions.

From a practical standpoint it would be much better also to give the percentage of spurs setting rather than, or in addition to, the per cent of blossoms because the orchardist usually wants only one fruit to a spur and may even go to the trouble of thinning all spurs with more than one fruit. The necessary percentage of spurs setting fruit to give a satisfactory crop under different percentages of spurs blossoming has not been worked out in detail and of course depends upon the size and condition of the tree, the size of the fruit of the variety in question and other factors. It is safe to say that most orchardists want their trees to bear as large crops as possible without the devitalizing effects of overbearing.

The choosing of checks or the determining of the normal set is a matter of great importance in pollination work. This is especially true in working with varieties that are weakly compatible. Thus, in hand pollination work where vigorous spurs are chosen for emasculation the same care should be used in selecting spurs for selfing or for open pollination. It is evidently undesirable to compare the set on vigorous spurs with spurs taken at random or with all the spurs on a limb or a tree. The normal set obtained by counting all blossoms and fruit on a large limb cannot be compared directly with the percentage set on spurs selected for their great vigor. The nearer the checks are to the blossoms with which they are compared the better. Great care should be exercised to have the checks strictly comparable since at best there are abundant possibilities of error.

In all pollination work it is important to check the viability of the pollen used by getting the percentage germination on artificial media. There is undoubtedly much to learn concerning the most suitable media to be used and the factors of temperature and concentration on pollen growth as well as varietal variation. Nevertheless in the work that has been done on pollination and pollen germination there has been a sufficiently high correlation between the ability of a variety to pollinate others satisfactorily and its viability as shown in germination tests to make such tests worth while.

### SUMMARY

The data on pollination experiments with the Baldwin apple is a good illustration of the fact that self-fruitfulness is not a fixed character in that

variety or any variety but varies greatly according to differences in environmental or other factors.

Pollination is only one of many factors which may be limiting to the setting of fruit. The exact effect of these factors is so little understood that their influence is very difficult to evaluate.

In studying pollination problems the experimenter should make all factors which influence fruit setting as favorable as possible so that no one will be limiting and observed differences will be due largely to differences in pollination. This is extremely difficult because of our inadequate knowledge of how these factors operate.

Practically all methods used in pollination work are open to criticism because they introduce unnatural factors, the effect of which we cannot evaluate.

A large part of the apparent confusion in the results of pollination experiments is due to the incomplete presentation of data which make it very difficult to interpret.

In presenting data information on the following points would be pertinent:

A. A statement of the condition of the trees used both as pistillate parents and sources of pollen and the previous treatment they have received, including: (1) planting distances; (2) whether the trees used are in sod or are cultivated; (3) the vigor of the trees as indicated by terminal, growth and color of the foliage; (4) the age of the trees in question; (5) the fertilizers applied to the trees, especially nitrates; (6) the amount and nature of any pruning that the trees have received within two years of the time of the experiment; (7) the amount of the crop of the preceding season; (8) the amount of bloom in the preceding season, and (9) the behavior of the tree the following season.

B. A full description of the method used, including: (1) the method used in selection of spurs for pollination; (2) the number of flowers pollinated per spur and their position on the spur; (3) the exact method of emasculation if flowers are emasculated; (4) the nature of the bags used and the length of time the flowers remained covered; (5) the source and treatment of the pollen used and how many trees furnished pollen and (6) in case the trees are caged with bees the stage of bloom on the caged tree when foreign pollen was introduced.

C. A statement of the results, including: (1) the per cent of spurs or growing points blooming; (2) the number of spurs pollinated; (3) the number of flowers pollinated; (4) the number of fruits set both before and after the June drop, the latter is of the most importance; (5) the number of spurs setting fruit, and (6) the percentage of pollen germinating on artificial media.

#### ACKNOWLEDGMENTS

The writer is indebted to Dr. A. J. Heinicke and Dr. F. S. Howlett for suggestions and criticism; and to Dr. E. L. Overholser of the University of

California, Dr. F. S. Howlett of the Ohio Agr. Exp. Sta. at Wooster, Ohio, and A. L. Pierstorff and R. Snyder at Cornell for the use of unpublished data.

#### LITERATURE CITED

1. Alderman, W. H. Experimental work on self-sterility of the apple. *Proc. Amer. Hort. Sci.* **1917**: 94-101.
2. Auchter, E. C., and Schrader, A. L. Cross fertilization of the Arkansas (Mammoth Black Twig) apple. *Proc. Amer. Soc. Hort. Sci.* **1925**: 96-105.
3. Chandler, W. H. Fruit growing. Houghton, Mifflin Co. 1925.
4. Ewert, R., 1906. Blütenbiologie und Tragbarkeit unserer Obstbäume. *Landw. Jahrb.* **35**: 259-287.
5. Gowen, J. W., 1920. Self-sterility and cross-sterility in the apple. *Me. Agr. Exp. Sta. Bull.* 287.
6. Heinicke, A. J., 1917. Factors influencing the abscission of flowers and partially developed fruits of the apple (*Pyrus Malus* L.). *Cornell Agr. Exp. Sta. Bull.* 393.
7. ———— 1927. Some factors to be considered in the practical application of sterility studies of fruits. *Mem. Hort. Soc. N. Y.* **3**:
8. Hendrickson, A. H., 1916. The common honey bee as an agent in prune pollination. *Agr. Exp. Sta. Bull.* 274.
9. Lewis, C. I., and Vincent, C. C., 1909. Pollination of the apple. *Ore. Agr. Exp. Sta. Bull.* 104.
10. MacDaniels, L. H. Pollination studies with certain New York State apple varieties. *Proc. Amer. Hort. Sci.* **1925**: 87-96.
11. Morris, O. M., 1921. Studies in apple pollination. *Wash. Agr. Exp. Sta. Bull.* 163.
12. Overholser, E. L., 1927. Apple pollination studies in California. *Mem. Hort. Soc. N. Y.* **3**:
13. Sax, K., 1922. Sterility relationship in Maine apple varieties. *Maine Agr. Exp. Sta. Bull.* 307.



# APPLE POLLINATION STUDIES IN CALIFORNIA

E. L. OVERHOLSER

*University of California*

Considerable study has been devoted to the pollination requirements of apple varieties by workers in the United States. Lewis and Vincent (1909), Auchter (1922), Gowen (1920), Morris (1921), Waugh (1898), Sax (1922), Vincent (1920), Wicks (1918), Crandall (1922), Powell (1902), Waite (1899), MacDaniels (1925) and others. Nevertheless it was of interest to conduct such studies in California, since the data of these workers were not always in agreement, and there is evidence that the pollination requirements of a single variety may vary when grown in different sections of the country (Tufts and Philp, 1923), and there are several varieties grown in California that had not previously received much attention.

The principal varieties grown in the two largest apple producing centers of California are the Yellow Newtown and the Yellow Bellflower in the Pajaro Valley, and the Gravenstein in the Sebastopol section. Many growers have observed that while these varieties generally bloomed profusely, the Yellow Bellflower and the Gravenstein in particular did not always set and develop commercial crops. On the other hand, occasionally certain growers cited instances where these varieties when planted in blocks as a single variety frequently set fair crops of fruit. In this connection it can be noted that in the coastal counties of California the blooming period of apples is relatively long, being sometimes six or eight weeks in duration. In the Mississippi Valley and the northeast the normal blooming period is generally not over a week to ten days. Furthermore, under California conditions, weather favorable for insect activity is more likely to prevail during a relatively greater part of the blooming period, hence it would appear that in normal seasons lack of proper varieties for cross-pollination would be less serious in these districts of California.

Because of this fact, although several years' data indicated the advisability of cross-pollination, there was some hesitancy in publishing the results until further field observations could be made.

During the spring of 1925, however, the set of Gravenstein apples throughout the Sebastopol section was exceptionally light as a result of unfavorable weather conditions for cross-pollination throughout the greater part of the blooming period. With the cooperation of the Sonoma County Farm Advisor's office several visits were made to a number of orchards, particularly those few that had a commercial crop.

It was apparent that only those orchards with varieties interplanted for cross-pollination were bearing crops. Furthermore, those varieties which the data indicated were satisfactory sorts for the cross-pollination of the

Gravenstein had resulted in a set, and those varieties, which on the basis of the data were unsatisfactory, had not resulted in a crop of Gravenstein apples. Furthermore, field observations in the Watsonville district also substantiated the data obtained.

### OBJECTS OF THE INVESTIGATION

The studies were chiefly made to determine the pollination requirements of the Yellow Newtown, Yellow Bellflower and Gravenstein.

Attempts were made to find satisfactory varieties for cross-pollination of these three varieties. The reciprocal cross with the pollinating sorts, together with the self-fruitfulness or self-unfruitfulness of the pollinating varieties was also studied.

The value of bees in effecting cross-pollination was demonstrated; the relative time of blooming, the abundance of pollen production, and the viability of the pollen of the more satisfactory varieties were noted.

The work was conducted over a period of five years in the vicinities of Watsonville and Sebastopol.\* All the trees were in a healthy, well-kept condition. The trees in Watsonville were about 20 years old and in Sebastopol the trees were from approximately 10 to 25 years old. In Sebastopol the varieties employed were the Gravenstein, Delicious, Jonathan, Esopus (Spitzenburg), Baldwin, Tompkins King, Yellow Bellflower and Rhode Island Greening; in Watsonville, the Yellow Newtown, Yellow Bellflower, White Winter Pearmain and Red Pearmain.

The two principal phases of the work were, first, to demonstrate the value of bees in pollination and, second, to study the pollination requirements by hand-pollinations.

### THE USE OF BEES IN POLLINATION

The blossom of the apple, like most commercial deciduous fruits is entomophilous, and supposedly requires insects, principally bees, for pollen transmission and pollination. Waugh (1898) and Lewis and Vincent (1909), by the use of microscope slides coated with vaseline and glycerine placed in the orchard, found that the pollen was not carried any great distance, nor in any abundance. Lewis and Vincent found that when the blossoms on a seven-year-old apple tree were emasculated, that only five fruits set. Apparently no pollen was carried by wind from a profusely blooming tree twenty feet away, and the insects were not attracted because only the stigmas of the flowers remained.

Hendrickson (1916) found that insects may be necessary for the application of pollen with even self-fruitful varieties of plums, and Alderman (1918) found that the placing of bees in an orchard increased the percentage of apple flowers setting fruit. More recently Auchter (1924) has found this to be true in Maryland.

---

\* In the Watsonville district the writer is under obligation to Mr. C. J. Rodgers for the use of orchard trees; and in the Sebastopol section to Mr. Wm. Hotle and Mr. Henry Elphick for the use of orchard trees and to the Sonoma County Farm Advisors for their cooperation.

With Hendrickson's work on plums in mind a large tent was erected enclosing two trees, one a Yellow Newtown and the other a Yellow Bellflower. Smaller tents were constructed to enclose, in one case a single Yellow Newtown tree, and in another case a single Yellow Bellflower tree. The tents were built of light redwood and covered with white mosquito-bar. Care was taken to avoid any openings through which insects might pass. Under each tent was placed a hive of bees to insure pollination.

As checks, selected limbs of other trees of the two varieties were enclosed in mosquito-bar netting to exclude all insects. The remaining limbs upon the trees so utilized were left exposed and untreated.

The percentage of set of fruit on the trees and branches enclosed by mosquito-bar was determined by counting the total number of blossoms upon certain branches. The number of flowers on each branch was recorded upon a manila tag attached to the base of each branch. Approximately two months later, or after a set of fruit should have resulted, count of the fruits was made and recorded. By comparison of the two counts the percentage of flowers which set fruit was obtained. These data were compared with the "normal set" of the particular variety in the orchard. Approximately five thousand blossoms representing the total number on selected branches on six representative trees distributed throughout the orchard were counted each year and later the fruits which developed were counted. Thus the "normal set" for the season was obtained.

### EXPERIMENTAL DATA

The data obtained during 1919, when the trees had a light to moderate bloom, in the study of the value of bees in cross-pollination are shown in TABLES 1 to 4.

TABLE 1

THE SET OF YELLOW NEWTOWN AND YELLOW BELLFLOWER CROSS-POLLINATED BY BEES (1919)

VARIETY CROSS-POLLINATED	NUMBER BLOSSOMS COUNTED	NUMBER APPLES SET	PER CENT OF SET
Newtown pollinated by Bellflower.....	1,794	918	51.50
Bellflower pollinated by Newtown.....	2,709	108	4.31

TABLE 2

THE SET UPON SINGLE TREES TENTED AND SELF-POLLINATED BY BEES

VARIETY SELF-POLLINATED	NUMBER BLOSSOMS COUNTED	NUMBER APPLES SET	PER CENT OF SET
Newtown.....	2,046	381	18.62
Bellflower.....	2,531	39	1.53

The data in TABLES 1 and 2 when compared with those in TABLE 3 show the value of bees in effecting pollination. The data in TABLE 3 indicate the necessity of insects in effecting a set of fruit. When insects were excluded almost no fruit resulted, while when bees were placed in the tents the set was considerable. In the case of Yellow Newtown cross-pollinated by bees with Yellow Bellflower over 50 per cent of the flowers set fruit.

TABLES 1 and 2 also show the value of cross-pollination as contrasted with self-pollination. TABLE 2 indicates that the Yellow Newtown is largely self-fruitful, but notwithstanding the high set of 19 per cent when self-pollinated, the set was greatly increased when cross-pollinated with Yellow Bellflower to 51.5 per cent. This is in contrast to the normal set of about 30 per cent with open pollination shown in TABLE 4.

TABLE 3  
THE SET UPON BRANCHES WITH INSECTS EXCLUDED BY MOSQUITO-BAR (1919).

VARIETY	NUMBER BLOSSOMS COUNTED	NUMBER APPLES SET	PER CENT OF SET
Newtown.....	1,768	5	0.28
Bellflower.....	958	3	0.31

TABLE 4  
THE NORMAL SET OF YELLOW NEWTOWN AND YELLOW BELLFLOWER APPLES (1919).

VARIETY	NUMBER BLOSSOMS COUNTED	NUMBER APPLES SET	PER CENT OF SET
Newtown.....	1,018	305	29.96
Bellflower.....	481	77	16.00

On the other hand the Yellow Bellflower is almost self-unfruitful since when self-pollinated by bees the set was less than two per cent (TABLE 2). While the set of Yellow Bellflower was increased when cross-pollinated by bees with Yellow Newtown, the percentage of set thus resulting was only slightly over four per cent (TABLE 1) as contrasted with the normal set of 16.0 per cent resulting from open pollination in the orchard (TABLE 4). This would indicate that while Yellow Newtown pistils and Yellow Bellflower pollen are cross-compatible, the Yellow Bellflower pistils and the Yellow Newtown pollen may be more or less cross-incompatible. In this connection, Hooper (1913) obtained evidence indicating that while Bramley pistils and Cox Orange pollen were cross-compatible, Cox Orange pistils and Bramley pollen were cross-incompatible.

The failure to set fruit when insects were excluded was tested again in 1920. In 1919 the flowers from which insects were entirely excluded were upon branches covered by mosquito-bar while the rest of the tree was exposed to the visits of insects and hence could be cross-pollinated.



Ewert (1906, 1907, 1909) appeared to find that parthenocarpic fruit, such as might develop without pollination, was at a disadvantage on the tree in competition with fruits containing developing seeds, such as might result from cross-pollination. It was, therefore, considered that failure to set fruit on the branches from which insects were excluded might have resulted from this unfavorable competition.

Hence, a single entire tree of the Yellow Newtown and the Yellow Bellflower were separately covered with mosquito-bar tents to exclude insects. In addition at random on ten trees each 2,371 blossoms of Yellow Newtown and 2,418 blossoms of Yellow Bellflower were covered with No. 10 manila bags just before the petals had expanded sufficiently to expose the pistils and stamens. Occasional flowers which were so far advanced as to have exposed the pistils and stamens were removed. After the petals had fallen, the paper bags were removed. Not a single fruit set either under the tents or in the bags notwithstanding the fact that the Yellow Newtown is self-fruitful. These data definitely indicate that these varieties of apples under the conditions of the experiment will not set fruit unless the pollen is actually applied to the pistils by insects or hand. This work is not in agreement with the work of Auchter (1922) who did not find any greater percentage of apple blossoms set fruit when he applied the pollen with a brush than when bagged blossoms were unmolested.

#### HAND POLLINATIONS

To prevent possible chance pollination of the stigmatic surfaces with pollen of the same flower, the blossoms employed for the hand pollination experiments were emasculated after the flowers had expanded appreciably, but before the overlapping petals had parted and the stigmatic surfaces were exposed or before the anthers had ruptured.

The emasculation, it was found, could be most expeditiously conducted using the nails of the thumb and second finger to cut through the floral envelope, removing the stamens together with the petals and a part of the calyxlobes, leaving only the stigmas. All flowers too far advanced or too immature or weak in their development were removed. This method with apples enabled from 1,500 to 2,000 emasculations per day to be made by a single worker and was more rapid than the use of scissors or forceps. The emasculated flowers were immediately covered by manila paper No. 10 sacks, and the date and number of emasculated flowers recorded on a tag affixed to the bag and on record sheets.

With the Yellow Newtown, Yellow Bellflower and Gravenstein varieties, from 20 to 25 trees in one to three orchards, were used for each season's pollination work. With the other varieties from 5 to 10 trees were used each season.

The flower clusters emasculated were on reasonably vigorous spurs, where it was expected conditions might be favorable for a certain percentage of flowers to set fruit. Between the time of emasculation and pollination

there was always some loss in the number of flowers due to injury at the time of emasculation, which escaped detection, or to some mechanical injury following emasculation. Such flowers were discarded at the time of pollination. An idea of the extent of this injury can be obtained by noting that out of about 72,800 flowers emasculated slightly over 62,000 flowers were in satisfactory condition for pollination, a loss in this manner of about 12 per cent.

Ewert (1907) states that the use of sacks in this manner subjects the enclosed spurs or portion of branch to unnatural conditions which may not be favorable for the setting of fruit. Heinicke (1917) found a larger percentage of Baldwin blossoms set fruit in translucent than in opaque sacks. He attributed this to the fact that the diffuse light in the translucent sacks permitted some photosynthetic activity, while nearly all light was excluded from the opaque sacks. Heinicke, however, in this experiment was troubled with aphid work. Aphids frequently cause apples to adhere which otherwise would absciss, and this may have affected the results. Furthermore, the spurs were sacked in the spring before the clusters of flowers had separated and were not removed until late summer. In the pollination work a week to ten days was generally the maximum period of time the blossoms and fruit were bagged. Alderman (1918) and Vincent (1920) found little difference in the percentage of flowers which set in bags and on trees under frames covered with thin cloth.

Since insects appeared necessary to effect pollination of apples and since Lewis and Vincent (1909) found insects did not visit emasculated flowers it was felt, in view of the opinions of Ewert and Heinicke, that it would be of interest to emasculate flowers and after tagging the spurs and branches leave them exposed without covering with the paper sacks. When the pistils became receptive the pollen was applied by hand, and the percentage of set compared with similarly treated blossoms enclosed in the paper sacks. Considerable time was saved in that the flowers did not have to be in such positions that a sack could be slipped over the spur or branch, and the time required to cover with sacks and tie the same was saved.

After two seasons' work, in 1920 and 1921 with approximately a total of 4,000 emasculated non-sacked blossoms of the Yellow Newtown and 4,200 of the Yellow Bellflower the method was, under the conditions existing and the varieties tested, considered unsatisfactory.

Apparently the emasculated blossoms tended to dry out very rapidly as a result of the wounded surfaces when not covered with sacks. The blossoms sacked did not dry out and the wounds healed. Furthermore, the blossoms that did not fall from excessive drying, appeared to have a shorter and less definite period when the stigmatic surfaces indicated a receptive condition of the pistils.

Sax (1922), however, seemed to find consistent results with uncovered emasculated apple flowers in Maine, and apparently considered the method satisfactory.

## COLLECTION OF POLLEN

In obtaining a supply of pollen the advanced but as yet unopened flower buds of each variety were collected from the trees a day or so before the dried pollen was needed. The unopened anthers were removed with forceps from the rest of the floral parts and deposited in Petri dishes with the lids partially raised so as to permit aeration and slight air circulation to aid in the drying. The anthers were dried in reduced sunlight, or occasionally during cloudy weather in an incubator at a temperature of 27° to 30° C. When dry the anthers and any escaped pollen were placed in small glass vials and kept loosely plugged with cotton in a dry place until ready for use.

## APPLICATION OF POLLEN TO PISTILS

The flowers were hand pollinated from one to three days after emasculation. Under the conditions of the experiments the stigmas were found to be in a receptive condition after this interval of time. It is a common practice to use a camel's hair brush in dusting the stigmas with pollen, but it was found that better application of the pollen with greater certainty was obtained by using the end of the cork stopper of the glass vial. By shaking the vial and occasionally breaking up the anthers with a pencil the pollen adhered to the stopper as a fine coating of yellow powder. The stopper was then removed and the stigmatic surface of each pistil was coated with pollen by simply touching the stigma with the pollen-covered end of the cork.

At the time of pollination the number of blossoms actually pollinated was counted, the number noted on the tags and record sheets, and the sacks were again placed over the clusters. When after the "June drop" the fruits were sufficiently advanced to determine the number that would continue to develop, the sacks were permanently removed and a count made of the young fruits. This count of apples was compared with the number of blossoms originally pollinated and the percentage of set determined. The percentage of set resulting from hand pollinations was always compared with the "normal set" of fruits untreated and exposed to orchard conditions and the visits of insects.

The viability of each lot of pollen employed in the hand pollinations was determined by germination tests in ten to twelve per cent cane sugar solutions.

## EXPERIMENTAL DATA

In the presentation of the data each of the more important varieties and crosses made are given in separate tables, and the data discussed under the variety heading.

*Yellow Newtown.* The data obtained with the Yellow Newtown are given in TABLE 5. The average set for three years when self-pollinated was about twelve per cent, as contrasted to the normal set during the same period

TABLE 5  
YELLOW NEWTOWN HAND POLLINATION EXPERIMENTS (WATSONVILLE)

POLLEN VARIETY	YEARS INCLUSIVE	NUMBER FLOWERS POLLINATED	NUMBER FRUITS SET	PER CENT SET	PER CENT NORMAL SET	PER CENT POLLEN GERM.
Selfed.....	1919-21	3,390	434	12.22	19.56	70
Yellow Bellflower.....	1919-21	2,797	1,123	30.76	19.56	66
Red Pearmain.....	1920-21	827	120	12.81	14.41	65
W. W. Pearmain.....	1920-21	1,103	222	18.93	14.41	75

of 19.5 per cent, which indicated the variety to be largely self-fruitful. This agrees with the work of Lewis and Vincent (1909) in Oregon. The work of Morris (1921) in Washington and Vincent (1920) in Idaho indicated the Newtown to be largely self-unfruitful. These workers, however, simply covered the blossoms and did not actually apply the pollen and the data previously presented indicate that such a method may not be a satisfactory one for determining self-unfruitfulness.

The Yellow Bellflower upon the basis of three years' work appeared to be a satisfactory pollinator for the Yellow Newtown, giving a set of nearly 31 per cent as contrasted to the normal set of 19.5 per cent. While Red Pearmain pollen appeared to be cross-compatible upon Yellow Newtown stigmas it possessed no especial merit as a pollinating sort, since it gave a percentage of set less than the normal set. The same was essentially true of the White Winter Pearmain although it gave a somewhat higher set than the normal set for the two years and might be considered as satisfactory for a pollinator of the Yellow Newtown.

*Yellow Bellflower.* The data in TABLE 6 with 4,328 self-pollinations

TABLE 6  
YELLOW BELLFLOWER POLLINATION EXPERIMENTS (WATSONVILLE AND SEBASTOPOL)

POLLEN VARIETY	YEARS INCLUSIVE	NUMBER FLOWERS POLLINATED	NUMBER FRUITS SET	PER CENT SET	PER CENT NORMAL SET	PER CENT POLLEN GERM.
Selfed.....	1919-23	4,328	14	0.07	8.5	74
Yellow Newtown.....	1919-23	4,445	133	4.29	8.5	79
Red Pearmain.....	1920-21	1,080	8	0.77	6.6	67
W. W. Pearmain.....	1920-21	1,086	6	0.85	6.6	72
Gravenstein.....	1922-23	899	0	0.00	6.6	25
Jonathan.....	1922	736	6	0.08	11.4	75
Tompkins King.....	1922-23	883	0	0.00	6.6	22
Esopus.....	1922	1,037	0	0.00	11.4	90
R. I. Greening.....	1922	965	0	0.00	11.4	40
Delicious.....	1923	163	5	3.00	1.9	50



show that the Yellow Bellflower is self-unfruitful. Furthermore, the only variety which appeared to give a set greater than the normal set was the Delicious. The number of blossoms employed with Delicious pollen, however, was small, 163, and the cross was made only one season. In addition, while the set with Delicious of 3.0 per cent was greater, the normal set was relatively small, 1.9 per cent. When the Yellow Bellflower was cross-pollinated with Yellow Newtown the average set for three years was a little over 4.0 per cent as contrasted to an average normal set of 8.5 during the same period.

The Red Pearmain with 1,080 crosses, W. W. Pearmain with 1,086 crosses, Gravenstein with 899 crosses, and Tompkins King with 883 crosses failed during two seasons' work to give an appreciable set when used to cross-pollinate the Yellow Bellflower. With one season's work only the Jonathan, with 736 crosses, the Esopus (Spitzenburg), with 1,037 crosses, and the Rhode Island Greening, with 965 crosses also failed to result in any set of Yellow Bellflower fruits. While the data cannot be taken as showing definite cross-incompatibility, they suggest this to be true in the case of Gravenstein, Tompkins King, Esopus, and Rhode Island Greening.

The pollen germination of the Gravenstein and the Tompkins King was unsatisfactorily low.

*Gravenstein.* The data in TABLE 7 show as the result of three years' work and 3,593 self-pollinations that the Gravenstein is self-unfruitful. This agrees with the work of Lewis and Vincent (1909) in Oregon; and Powell (1902) in Delaware. Morris (1921) in Washington and Vincent (1920) in Idaho, however, found that when the Gravenstein blossoms were bagged and not actually hand pollinated that the percentages setting fruit were 5.1 and 3.5 respectively.

The Delicious proved to be the best pollinator for the Gravenstein, during three years' work as a result of 3,031 crossings giving an average set of 9.3

TABLE 7  
GRAVENSTEIN. POLLINATION EXPERIMENTS (SEBASTOPOL)

POLLEN VARIETY	YEARS INCLUSIVE	NUMBER FLOWERS POLLINATED	NUMBER FRUITS SET	PER CENT SET	PER CENT NORMAL SET	PER CENT POLLEN GERM.
Selfed.....	1921-23	3,593	3	0.09	6.75	14
Esopus.....	1921-23	2,748	62	2.60	6.75	81
Delicious.....	1921-23	3,031	305	9.30	6.75	78
Jonathan.....	1921-23	2,395	43	2.34	6.75	72
R. I. Greening.....	1922-23	1,239	0	0.00	6.63	50
Yellow Newtown....	1922-23	1,334	67	6.20	6.63	73
Yellow Bellflower....	1922-23	1,715	5	0.30	6.63	78
Tompkins King.....	1922-23	1,816	0	0.00	6.63	30
Baldwin.....	1923	890	0	0.00	9.10	25

per cent as contrasted to the normal set of 6.75 per cent. Morris (1921) crossed 99 Gravenstein blossoms with Delicious pollen and found 16 per cent of the blossoms set fruit. Another satisfactory pollinator was the Yellow Newtown giving a set comparable to the normal set. Morris (1921) crossed 34 Gravenstein blossoms with Jonathan pollen and found about 18 per cent set fruit, but with 57 Gravenstein blossoms crossed with Esopus none of them set fruit. Vincent (1915) also found the Jonathan was cross-fruitful upon the Gravenstein. The Rhode Island Greening, Yellow Bellflower, Tompkins King, and Baldwin appeared to be cross-incompatible with Gravenstein.

The Gravenstein did not produce abundant pollen and the pollen lacked viability as shown by the low average germination percentage of 14.0. It would appear that the self-unfruitfulness and the cross-incompatibility with other varieties might be partially due to this. With the pollen of the Tompkins King and the Baldwin used in cross-pollinating the Gravenstein the germination was also unsatisfactorily low, 30.0 and 25 per cent respectively.

*White Winter Pearmain.* The two years' data in TABLE 8 show the White Winter Pearmain to be self-unfruitful. This agrees with the work of Auchter (1922), Morris (1921), and Vincent (1920). The best pollinator of the White Winter Pearmain appeared to be the Red Pearmain, although the set resulting was about one-half that of the normal set. The Yellow Bellflower and the Yellow Newtown when used as pollinators were both inter-fruitful with the White Winter Pearmain.

TABLE 8  
WHITE WINTER PEARMAIN POLLINATION EXPERIMENTS (WATSONVILLE)

POLLEN VARIETY	YEARS INCLUSIVE	NUMBER FLOWERS POLLINATED	NUMBER FRUITS SET	PER CENT SET	PER CENT NORMAL SET	PER CENT POLLEN GERM.
Selfed.....	1920-21	940	0	0.00	11.32	72
Yellow Bellflower.....	1920-21	771	15	2.12	11.32	71
Yellow Newtown.....	1920-21	943	13	1.71	11.32	78
Red Pearmain.....	1920-21	1,479	93	5.58	11.32	63

*Tompkins King.* The data from two years' work as shown in TABLE 9 indicate that the Tompkins King is self-unfruitful as reported by Lewis and Vincent (1909), and Morris (1921). Auchter, however, by employing blossoms covered but not actually hand pollinated reported that in Maryland the Tompkins King was self-fruitful. The germination tests indicated a somewhat low viability of the Tompkins King pollen and markedly low viability or imperfect pollen for the Gravenstein.

*Red Pearmain.* The data in TABLE 10 indicate that the Red Pearmain is self-unfruitful, and that the pollen of Yellow Bellflower, Yellow Newtown and White Winter Pearmain is cross-compatible with the stigmas of

TABLE 9  
TOMPKINS KING POLLINATION EXPERIMENTS (SEBASTOPOL)

POLLEN VARIETY	YEARS INCLUSIVE	NUMBER FLOWERS POLLINATED	NUMBER FRUITS SET	PER CENT SET	PER CENT NORMAL SET	PER CENT POLLEN GERM.
Selfed.....	1922-23	1,990	0	0.0	27.6	30
Gravenstein.....	1922-23	1,349	0	0.0	27.6	8
R. I. Greening.....	1922-23	853	0	0.0	27.6	67
Yellow Bellflower.....	1922-23	1,411	30	1.7	27.6	78
Jonathan.....	1922-23	1,072	190	14.1	27.6	73
Yellow Newton.....	1923	359	0	0.0	18.9	70

Red Pearmain. While the sets of Red Pearmain pollinated with Yellow Bellflower and Yellow Newtown were less than the normal set; that resulting from White Winter Pearmain pollen was in excess of the normal set.

TABLE 10  
RED PEARMAIN POLLINATION EXPERIMENTS (WATSONVILLE)

POLLEN VARIETY	YEARS INCLUSIVE	NUMBER FLOWERS POLLINATED	NUMBER FRUITS SET	PER CENT SET	PER CENT NORMAL SET	PER CENT POLLEN GERM.
Selfed.....	1920-21	306	1	0.19	7.16	63
Yellow Bellflower.....	1920-21	893	33	3.57	7.16	70
Yellow Newtown.....	1920-21	890	39	2.65	7.16	67
W. W. Pearmain.....	1921	676	59	8.89	7.22	74

*Esopus (Spitzenburg)*. The data of three years' in TABLE 11 indicate the *Esopus (Spitzenburg)* is self-fruitful. This agrees with the work of Lewis and Vincent (1909). Morris (1921) and Waugh (1898) likewise obtained data which showed a tendency for the variety to be self-fruitful. Vincent (1920) and Waite (1899), however, appeared to find it self-unfruitful under Idaho and New York conditions, respectively. The pollen of Gravenstein, as a result of three seasons' work, was shown to be cross-incompatible upon *Esopus (Spitzenburg)* pistils. The Gravenstein pollen, however, gave a low germination test. Both the Jonathan and the Delicious pollen gave exceptionally high sets, 55.5 and 36.4 per cent, respectively, as contrasted to the normal set of 11.3. Lewis and Vincent found the pollen of Jonathan to be highly compatible upon the stigmas of *Esopus (Spitzenburg)* and Norris found this was true for both Jonathan and Delicious pollen upon *Esopus (Spitzenburg)* pistils.

*Additional Varieties*. The Rhode Island Greening, Jonathan, Delicious, and Baldwin were in each case self-pollinated and cross-pollinated with Gravenstein. The data are given in TABLE 12. The work of two years with

TABLE 11  
ESOPUS (SPITZENBERG) POLLINATION EXPERIMENTS (SEBASTOPOL)

POLLEN VARIETY	YEARS INCLUSIVE	NUMBER FLOWERS POLLINATED	NUMBER FRUITS SET	PER CENT SET	PER CENT NORMAL SET	PER CENT POLLEN GERM.
Selfed.....	1921-23	1,037	67	4.4	20.5	85
Gravenstein.....	1921-23	1,004	2	0.1	20.5	12
Jonathan.....	1921	632	332	55.5	11.3	70
Delicious.....	1921	730	266	36.4	11.3	85

Rhode Island Greening, Jonathan, and Delicious indicate that these varieties are self-unfruitful.

The work of Crandall (1922), Morris (1921), and Vincent (1920) with the Delicious agrees with these data. The data with the Jonathan agree with the findings of Lewis and Vincent in Oregon and Morris in Washing-

TABLE 12  
ADDITIONAL POLLINATION EXPERIMENTS

VARIETY POLLINATED	POLLEN VARIETY	YEARS INCLUSIVE	NUMBER FLOWERS POLLINATED	NUMBER FRUITS SET	PER CENT SET	PER CENT NORMAL SET	PER CENT POLLEN GERM.
R. I. Greening	Selfed.....	1922-23	582	0	0.0	9.8	43
R. I. Greening	Gravenstein..	1922-23	449	0	0.0	9.8	12
Jonathan.....	Selfed.....	1921-23	600	1	0.4	28.4	70
Johnathan....	Gravenstein..	1922-23	461	0	0.0	28.4	12
Delicious.....	Selfed.....	1922-23	426	0	0.0	22.0	57
Delicious.....	Gravenstein..	1922	150	0	0.0	9.1	15
Baldwin.....	Selfed.....	1923	209	15	7.1	19.8	15
Baldwin.....	Gravenstein..	1923	99	0	0.0	19.8	10

ton, but Vincent in Idaho, and Wicks (1918) in Arkansas seemed to find the Jonathan partly self-fruitful. Crandall (1922), Gowen (1920), Lewis and Vincent, Vincent, Waite (1899), Waugh (1898), and Sax (1922) found the Rhode Island to be self-unfruitful, although Morris found it partially self-fruitful.

The data in TABLE 12 indicated the Baldwin was self-fruitful. This agrees with the work of Gowen and Lewis and Vincent. Morris, Auchter (1922) and MacDaniels (1925) also found the Baldwin partly self-fruitful. Waugh and Sax, however, found the Baldwin self-unfruitful, although Waugh simply bagged the blossoms without actually applying the pollen.

The pollen of Gravenstein appeared to be cross-incompatible with each of the four varieties. The Gravenstein pollen viability was low, although this was also true of the Baldwin pollen which gave a set upon Baldwin stigmas of about 7 per cent.



## DISCUSSION AND SUMMARY

1. The use of bees as a means of effecting pollination in an apple orchard greatly increased the set of fruit when contrasted to the normal set.

2. Cross-pollination with effective varieties of even a self-fruitful variety as the Yellow Newtown increased the set of fruit.

3. The actual application of the pollen by insects or by hand to insure pollination of Yellow Newtown and Yellow Bellflower blossoms seemed necessary.

4. In the case of these two varieties, the method of bagging or enclosing blossoms with mosquito-bar to determine self-compatibility or self-incompatibility seemed unreliable. No fruits set with even a self-fertile variety when blossoms were bagged, or single branches and an entire tree were covered with mosquito-bar.

5. In the case of emasculated Yellow Newtown or Yellow Bellflower blossoms it seemed advisable to enclose the same in sacks to lessen the drying out of the wounded remaining floral parts and to prolong the period of receptivity. There was no evidence, however, to indicate the advisability of bagging to exclude chance pollination of emasculated flowers.

6. The average normal set of from one to four years under orchard conditions for the eleven varieties studied was 14 per cent. A rather high average set of fruit.

7. Under the conditions of the experiment the Yellow Newtown, Esopus (Spitzenburg) and Baldwin were self-fruitful.

8. The average set, of those varieties considered as self-fruitful, resulting from self-pollinations, was 10.6 per cent.

9. The Yellow Bellflower, Gravenstein, White Winter Pearmain, Tompkins King, Rhode Island Greening, Delicious, Red Pearmain, and Jonathan were considered self-unfruitful.

10. The average set of those varieties classed as self-unfruitful was 0.19 per cent.

11. The Gravenstein did not satisfactorily cross-pollinate any of the varieties tested. This, at least partially appeared to be the result of defective and non-viable pollen as determined by microscopic examination of the pollen grains and by germination tests.

12. The evidence is not conclusive, but the data indicate that the pollen of Tompkins King, Esopus, and Rhode Island Greening is cross-incompatible upon the stigmas of the Yellow Bellflower; that pollen of Rhode Island Greening, Tompkins King, and Baldwin is cross-incompatible upon the Gravenstein; and that pollen of Rhode Island Greening, and Yellow Newtown is cross-incompatible upon the stigmas of Tompkins King.

13. The Yellow Bellflower, Red Pearmain and the White Winter Pearmain were considered as good varieties to pollinate the Yellow Newtown; the Yellow Newtown and Delicious for the Yellow Bellflower and for the Gravenstein; the Red Pearmain for the White Winter Pearmain; the Jon-

athan for the Tompkins King; the White Winter Pearmain for the Red Pearmain; and the Jonathan and Delicious for the Esopus (Spitzenburg).

14. The average set of the varieties to which reference has just been made when cross-pollinated with the satisfactory pollinators was 18.7 per cent.

15. The pollen of certain varieties was considered as partially cross-compatible upon the stigmas of certain flowers but not sufficiently so for commercial cross-pollination plantings. This was true of Esopus and Jonathan pollen upon the stigmas of Gravenstein; the Yellow Bellflower and Yellow Newtown upon White Winter Pearmain; the Yellow Bellflower upon Tompkins King; and Yellow Bellflower and Yellow Newtown pollen upon Red Pearmain stigmas.

16. The average set resulting from pollination with these partially cross-compatible varieties was 2.4 per cent.

*Acknowledgments.* The writer wishes to express appreciation of help received from certain graduate students as follows: E. S. Yocco, 1919; H. E. Jacob, 1920-21; L. P. Latimer and R. H. Rawl, 1922-23. Drs. L. H. MacDaniels and A. J. Heinicke have very kindly read the manuscript and offered helpful suggestions.

#### BIBLIOGRAPHY

- Alderman, W. H., 1918. Experimental work on self-sterility of the apple. *Proc. Amer. Hort. Sci.* **1917**: 94-101.
- Auchter, E. C., 1922. Apple pollen and pollination studies in Maryland. *Proc. Am. Soc. Hort. Sci.* **18**: 51-80.
- Crandall, C. S., 1922. Results from self-pollination of apple flowers. *Proc. Amer. Soc. Hort. Sci.* **1921**: 95-100.
- Ewert, R., 1906. Blüten biologie und Trag-barkeit unserer Obstbäume. *Landw. Jahrb.* **35**: 259-287.
- 1907. Die Parthenokarpie oder Jungfernfruchtigkeit der Obstbäume, pp. 1-58.
- 1909. Neuere Untersuchungen über Parthenokarpie bei Obstbäumen und einigen anderen fruchttragenden Gewachsen. *Landw. Jahrb.* **38**: 767-839.
- Gowen, J. W., 1920. Self-sterility and cross-sterility in the apple. *Me. Agr. Exp. Sta. Bull.* **287**.
- Heinicke, A. J., 1917. Factors influencing the abscission of flowers and partially developed fruits of the apple (*Pyrus malus* L.). *Cornell Agr. Exp. Sta. Bull.* **393**.
- Hendrickson, A. H., 1916. The common honey bee as an agent in prune pollination. *Calif. Agr. Exp. Sta. Bull.* **274**.
- Hooper, C. H., 1913. The pollination of fruit trees and its bearing on planting. *Gard. Chron.* 3 ser. **54**: 393-34, 420.
- Lewis, C. I., and Vincent, C. C., 1909. Pollination of the apple. *Ore. Agr. Exp. Sta. Bull.* **104**.
- MacDaniels, L. H., 1926. Pollination studies with certain New York State apple varieties. *Proc. Amer. Hort. Sci.* **1925**: 87-96.
- Morris, O. M., 1921. Studies in apple pollination. *Wash. Agr. Exp. Bull.* **163**.
- Powell, G. H., 1902. The pollination of apples. *Del. Agr. Exp. Sta. Rept.* **13**: 112-16.
- Sax, K., 1922. Sterility relationship in Maine apple varieties. *Me. Agr. Exp. Sta. Bull.* **307**.
- Tufts, W. P., and Philp, Guy L., 1923. Pear pollination. *Calif. Agr. Exp. Sta. Bull.* **373**.
- Vincent, C. C., 1915. *Rept. of Dept. of Hort. in Bull.* **84**, Idaho Agr. Exp. Sta., p. 24.
- 1920. Results of pollination studies at Idaho Univ. *Better Fruit* **14**: 11-15.
- Waite, M. B., 1899. Pollination of pomaceous fruits. *U. S. D. A. Yearbook* (1898), **167-80**.
- Waugh, F. A., 1898. *Proc. Amer. Pomol. Soc.*, **25**: 86-94.
- Wicks, W. H., 1918. The effect of cross-pollination on the size, color, shape and quality of the apple. *Ark. Agr. Exp. Sta. Bull.* **143**.

# THE RESULTS OF CROSS-POLLINATION BETWEEN DIFFERENT VARIETIES OF APPLES, PEARS, PLUMS AND CHERRIES

RICHARD WELLINGTON

*New York Agricultural Experiment Station*

Although the New York Agricultural Experiment Station has carried on considerable work in the study of sterility and compatibility of fruits, especially the grape, more attention has been given in recent years to the production of new varieties. Lately, Dr. A. B. Stout has been cooperating with our Station in the study of fruit sterilities and compatibilities and has already published on some of his studies. In crossing, certain combinations are found to give fruit and good seed, while others give nothing. It is upon these findings that this report is based. Pollen germination was not always tested, but when pollen taken from the same source proves effective in one cross and not in another one has reason to form an opinion. Undoubtedly certain failures would be successes under different conditions, especially as we find certain crosses behave differently from year to year.

## APPLES

Over 100 apple varieties have been crossed and selfed at the Geneva Station since 1898. Some of these varieties were used in many crosses, and others in comparatively few. As our time is limited, only a few of the more important ones will be noted.

Arkansas was crossed in 1923 with high quality varieties, namely, Cortland, Cox Orange, Golden Delicious, McIntosh, Macoun, and Red Spy. All the crosses, with the exception of the Red Spy set fruits. Cortland and Golden Delicious crosses gave on the average about one good seed per fruit, Cox Orange and Macoun one good seed to three fruits, and McIntosh about three seeds to one fruit. The reciprocal cross with McIntosh gave from two to three seeds per fruit, but the reciprocal cross with Red Spy gave no fruit. Judging from one year's results, Red Spy and Arkansas are incompatible, but from the orchardist's standpoint it makes little difference as these two varieties are not grown commercially in the same region.

Baldwin has not been used as extensively in breeding work as its commercial value might warrant. In 1906, 36 clusters of flowers were selfed on two trees, and in June, 18 fruits were noted. Unfortunately the number of mature fruits was not recorded. Varieties crossed on the Baldwin that have given fruit and seed are Opalescent, Cortland, McIntosh, and Red Spy. Dr. Stout could add the Delicious to this list. Although Cortland and Opalescent pollen fertilized the Baldwin, Baldwin pollen failed when used on

Opalescent and Cortland. Cox Orange crossed on Baldwin also failed.

Ben Davis was selfed in 1906 and 1907, and although 53 flower clusters were covered only one fruit was noted in June. Possibly this fruit dropped without maturing. Esopus Spitzenburg, Green Newtown, Jonathan, McIntosh and Mother, when used as males on Ben Davis, produced fruit, while Blenheim gave no fruit and Gravenstein no fruit one year and one apple another year. Pollen of Sheldon, Clairgeau, and Beurré Bosc pears applied to Ben Davis gave no mature fruits, although three very small fruits were recorded in June in the Sheldon cross. Ben Davis pollen when used on Esopus Spitzenburg produced fruit but Ben Davis pollen caused no set on Boiken.

Cortland, a variety originated at the Geneva Station, has been crossed with numerous varieties. Male parents that have given fruit and seed when used on Cortland, are McIntosh, Green Newtown, Red Spy, and Crimson Beauty. Baldwin, as already noted, gave no set when used in 1924 as a male on this variety. Cortland has fertilized satisfactorily Arkansas, Baldwin, King David, McIntosh, Orenco, Northern and Red Spy, Rhode Island Greening, and Wealthy.

Cox Orange when used as a male produced fruit on Arkansas, Early McIntosh, McIntosh, Rhode Island Greening, and Red Spy. An average of over 13 seeds to the fruit were obtained from the Early McIntosh cross, while less than one good seed to three fruits were obtained in the Arkansas cross. Cox Orange pollen did not function when used on Baldwin and Sweet McIntosh. Both Early McIntosh and Golden Delicious, when used as males, caused Cox Orange to develop fruit and seeds.

Golden Delicious produced fruit when fertilized with Arkansas, King David, Red Spy, and Yellow Newtown. When used as a male, fruit was secured from Arkansas, Cox Orange, King David, Red Spy, Deacon Jones, and Delicious. Yellow Bellflower by Golden Delicious, however, gave no fruit.

Delicious has proved to be incompatible in many crosses, practically all varieties used as pollinators having failed. Varieties that have given no fruit when used as males are: Chenango, Gilliflower, Cortland, Early McIntosh, King David, McIntosh, Northern Spy, Red Spy, and Yellow Newtown. Although Delicious gave no fruit when selfed or when used on Chenango and Golden Delicious and no good seed on Yellow Newtown, it fertilized Deacon Jones, Baldwin (according to Stout), Gilliflower, McIntosh, Red Canada, Red Spy, and Winter Banana. Three of the Delicious crosses thus worked one way and not the other.

Jonathan when selfed has given no fruit, although its pollen germinated well. When used as a male on Ben Davis, Jersey Blue, and Rome fruit was obtained but on Sutton no fruit was secured. Jersey Blue, Rome, and Esopus Spitzenburg fertilized Jonathan satisfactorily.

McIntosh has proved to be an excellent parent and consequently has been used in many crosses. When selfed under paper bags this variety has



proved to be practically self-incompatible as 30 clusters gave only one fruit. Dr. Stout, on the other hand, obtained a fair set under a tent which also enclosed a hive of bees. Varieties which have been found to fertilize McIntosh are Anis Rose, Arkansas, Baldwin, Cortland, Cox Orange, Crimson Beauty, Delicious, Deacon Jones, Duchess of Oldenburg, Golden Delicious, Jersey Blue, King David, Lawver, Northern Spy, Primate, Rhode Island Greening (Dr. Stout's cross), Wealthy, Yellow Newtown, and Yellow Transparent. In fact only two varieties, namely, Orenco and King Seedling have given negative results. McIntosh pollen when used on Anis Rose, Arkansas, Baldwin, Cortland, Deacon Jones, Duchess of Oldenburg, Jersey Blue, Lawver, Orenco, Northern Spy, Red Spy, Wealthy, Yellow Transparent, and Zuzoff Winter also produced fruit. No fruit, however, was obtained when McIntosh was used on Delicious, Early McIntosh, King Seedling, Macoun, Primate, and San Jacinto. As the number of crosses were few, the work should be repeated before drawing definite conclusions.

Northern Spy and Red Spy, a probable sport, have also been used extensively in our breeding work. Selfed Northern Spy flowers have given no fruit, although its pollen germinated excellently. Northern Spy pollen when used on Deacon Jones, Green Newton, Macoun, McIntosh, Ralls, Rome and Wellington Bloomless has given fruit and seeds. Failures to set, however, were recorded when Northern Spy pollen was used on Delicious and Tompkins King with the exception of one fruit noted in July. Red Spy likewise gave no fruit when used on Delicious and no set on Arkansas and Rhode Island Greening. Red Spy proved compatible, however, when used on Deacon Jones, Cortland, Early McIntosh, Golden Delicious, and McIntosh. Cortland and McIntosh fertilized both the Northern Spy and Red Spy. Yellow Newtown was also used successfully on Northern Spy; and Cox Orange, Delicious, and Golden Delicious on Red Spy.

Rhode Island Greening pollen tested in 1903 gave a germination of 8 to 10 per cent and 45 bags used in covering clusters of flowers on three trees during the years of 1906 and 1907 gave a total of 11 fruits in June. Whether any of these fruits matured was not recorded. Varieties which have given fruit when used on Rhode Island Greening are Cortland, Cox Orange, Esopus Spitzenburg, King Seedling, Orenco, and Yellow Newtown. Red Spy, as a rule, failed to produce a set. Rhode Island Greening pollen failed when used two seasons on Esopus Spitzenburg, although the reciprocal cross as just noted proved successful.

There may be a question as to whether the negative data here recorded were not due to other causes than incompatibility. Inclement weather, causing delayed pollination, may have been a factor in some cases, and again the pollen may not have been in the best condition when applied. If the crosses had been repeated a set might have been obtained in a few cases; however, the data are presented so that they can be used to corroborate or to refute results obtained by others.

## PEARS

About 40 varieties of pears have been used in our breeding work but only a few will be mentioned.

Beurré D'Anjou failed to give seed when pollinated with Bartlett, Doyenné Boussock, Seckel, and Tyson; but, on the contrary, all these varieties, with the exception of Doyenné Boussock, set freely in the reciprocal crosses. Buerré d'Anjou also fertilized Beurré Bosc, Clairgeau, Kieffer, Phelps, and Pulteney.

Bartlett has been used in many crosses and has given seed when pollinated with the following varieties, namely, Beurré d'Anjou, Beurré Dumont, Beurré Bosc, Beurré Giffard, Clairgeau, Dana Hovey, Doyenné Boussock, Duchesse d'Angoulême, Dorset, Eureka, Glou Morceau, Pitmaston (1 seed), Sheldon, Tyson, and Winter Nelis. No seed, however, was obtained when Bartlett was pollinated with Alexander Lucas, Lawrence, Russet Bartlett, Seckel, and Worden Seckel. Seckel pollen was used three different years. Doyenné Boussock, on the other hand, fertilized the Bartlett one year but not for two years. Bartlett, as a male, was found to be compatible with Beurré Giffard, Beurré Dumont, Dana Hovey, Doyenné Boussock, Eureka, Josephine de Malines, Lincoln Coreless, P. Barry, Seckel, Tyson, and Winter Nelis and incompatible with Russet Bartlett. Also, seed was obtained when Bartlett pollen was used on Beurré d'Anjou and Beurré Bosc, but not enough crosses were made to make a definite statement as to their compatibilities. Russet Bartlett, a probable sport from Bartlett, set no fruit when selfed or when crossed with Bartlett and LeConte. Winter Nelis, however, proved to be a good pollinator for Russet Bartlett.

Beurré Bosc, as a female, was found to be compatible with Beurré d'Anjou, Buerré Giffard, Duchesse d'Angoulême, Dana Hovey, Kieffer, P. Barry, Seckel, and Tyson, and incompatible with Bartlett, Doyenné Boussock, and Pitmaston. Beurré Bosc, as a male, however, fertilized Bartlett, Doyenné Boussock, and Pitmaston as well as Dana Hovey, Beurré Dumont, Kieffer, Lincoln Coreless, P. Barry, Pitmaston, Phelps, Pulteney, Tyson and Winter Nelis, but not Flemish Beauty (one trial).

Kieffer has failed in no crosses attempted. Abundant seed was obtained when Bartlett, Beurré d'Anjou, Beurré Bosc, Flemish Beauty, Manning Elizabeth, Phelps, Pulteney, and Seckel were used as males. Kieffer fertilized Beurré Bosc and Flemish Beauty.

Many varieties have been found to fertilize the Seckel, although there is apparently a variation in the degree of compatibility. For example, Bartlett gave no set in 1923 and 1924 and a light set in 1925 and 1926 while other varieties have always given good sets. Compatible varieties, used as males, are Bartlett, Beurré d'Anjou, Beurré Dumont, Beurré Giffard, Beurré Bosc, Clairgeau, Dana Hovey, Doyenné Boussock, Durandau, Duchesse d'Angoulême, Gorham, Phelps, Pitmaston, Pulteney, Sheldon and Tyson. Seckel pollen was used for three years on Bartlett without any success and also for one year on Beurré d'Anjou, LeConte, Pitmaston, and Rankin. Seed, how-

ever, was obtained when Seckel was used to pollinate Beurré Bosc, Clairgeau, Kieffer, Lincoln Coreless, P. Barry, Pulteney, Tyson, and Winter Nelis.

Winter Nelis, although self-incompatible in one test, set freely when pollinated with Bartlett, Beurré Bosc, Durandeau, Lincoln Coreless, Russet Bartlett, Seckel, Sheldon, and Tyson. Pollen of Winter Nelis also functioned when used on Bartlett and Lincoln Coreless.

## PLUMS

Sixty-six varieties and many seedlings have been used in our breeding work with plums, but as in the case of the other fruits only a few will be mentioned at this time. Abundance, a *Prunus salicina*, fertilized the Wickson but gave no set on Ogon, Jefferson, Quackenboss, and Surprise. Thus, the inter-species crosses and one intra-species cross failed.

Agen set fruit when selfed and pollinated with sixteen other varieties of *Prunus domestica*. No failures were recorded. Agen pollen, however, gave, as a rule, a much poorer set and in crosses with Sannois and Transparent produced no seed. Usually no seed was obtained when crosses were made between *Prunus salicina* and *Prunus domestica*, although one seed was harvested in the cross Burbank by Imperial Epineuse. Under a tent which enclosed a tree of Burbank and Reine Claude and a colony of bees, Prof. L. H. MacDaniels of Cornell secured many seeds of Burbank and Reine Claude, both varieties being self-incompatible. The seed was turned over to the Geneva Station and only one doubtful tree was obtained.

Burbank crossed with five Japanese varieties set satisfactorily, but when used as a male on four European kinds no seed was secured. Burbank, however, caused the development of fruit when used on Marianna, Eldorado, and Santa Rosa.

Grand Duke when selfed gave no fruit, but when pollinated with nine *Prunus domestica*s set fruit. Thirteen *domestica*s pollinated with Grand Duke set fruit, but a *Prunus americana* pollinated with Grand Duke produced no fruit.

Imperial Epineuse when selfed gave no fruit but set fruit when pollinated with Agen, Drap d'Or, Grand Duke, Hand, Jefferson, Miller Superb, Mirabelle, Middleburg, Pond, and Yellow Egg. McLaughlin and Pearl, however, did not give a set but they were used only one season. Imperial Epineuse also gave a good set when used as a male on Agen, Freestone, Jefferson, Pond, and Yellow Egg; a small set on Grand Duke, Hand, Middleburg, and Reine des Mirabelles; and no set on Arch Duke, Golden Beauty, and Sannois.

## CHERRIES

Little need be said about cherries as several excellent papers have appeared on this fruit. Two new varieties, namely, Giant and Seneca, however, should be mentioned.

Giant is an excellent, firm-fleshed, black cherry and has proved to be a

good pollinator for Abundance, Lyons, Lambert, Montmorency, and Napoleon. Abundance, Lambert, Napoleon, and Windsor also pollinate the Giant, so we have a new variety of commercial possibilities to add to our cherry industry.

Seneca, a new, very early, black cherry, produces pollèn very profusely, and although it refused to set when selfed, it pollinated successfully Black Tartarian, Ida, Giant, Lyons, Napoleon, Republican, Schmidt, Windsor, and Yellow Spanish. In no cross has Seneca given any evidence of being incompatible.



# FIELD STUDIES OF THE POLLINATION REQUIREMENTS OF CERTAIN DECIDUOUS FRUITS UNDER CALIFORNIA CONDITIONS

W. P. TUFTS, A. H. HENDRICKSON and G. L. PHILP

*University of California*

Field tests designed to determine the sterility of various deciduous fruits have been conducted by the Pomology Division of the University of California each season for the past twelve years. The purpose of these investigations has been to secure information of immediate value and use to the fruit growers of California and also to gradually accumulate data upon which to base a more intimate and detailed study of the factors causing sterility in fruits.

Study of the pollination requirements of the almond, apricot, cherry (*Prunus avium*), peach, pear, and plum (*Prunus domestica* and *Prunus salicina*) has been made. Progress reports dealing with the almond,<sup>1</sup> cherry,<sup>2</sup> pear,<sup>3</sup> and plum<sup>4</sup> have been published. The conclusions reached have been based on rather comprehensive data collected through a period of years and under widely differing environmental conditions. In order to convey an idea of the scope of the work mention may be made of the fact that 150,000 almond, 375,000 cherry, 90,000 pear and 223,000 plum blossoms were actually emasculated and hand pollinated. This in addition to more than 825,000 blossoms used in estimating "normal sets."

In the course of all this work a technique has naturally developed which may in some respects differ from that normally employed in pollination studies. In collecting pollen for artificial crosses flowers were gathered just before they open. Immature blossoms and the first blooms do not contain as viable pollen as the more mature flowers which open later. The anthers were removed from the flowers and dried in petri dishes in the sunshine until thoroughly dry. The pollen was then stored in cotton stoppered vials until needed. In all the tests the pollen was artificially tested in a 12 per cent to 15 per cent cane sugar solution; germinations having been made in Van Teigham cells, using the hanging drop. The pollen is most easily applied to the pistils with either a cork or the tip of the finger.

In recent years the writers have discarded paper sacks for the protection of emasculated and pollinized flowers. We have found that the use of sacks is unnecessary as bees and other insects do not visit emasculated flowers, and that the chance of accidental pollination is so slight (generally less than one-

<sup>1</sup> Calif. Agri. Expt. Sta. Bull. 346.

<sup>2</sup> Calif. Agri. Expt. Sta. Bull. 385.

<sup>3</sup> Calif. Agri. Expt. Sta. Bull. 373.

<sup>4</sup> Calif. Agri. Expt. Sta. Bulls. 291, 310, 352.

tenth of one per cent) that it is negligible. The writers are of the opinion that the error introduced by injury to pistils which is caused by bagging and removal of sacks is greater than that caused by accidental pollination of unprotected pistils. There is much more danger from frost injury under sacks than when the emasculated flowers are exposed. Many more pollinations can be made in the same length of time which also is an advantage, not to mention the cost of the sacks and time lost in using them. In making crosses in

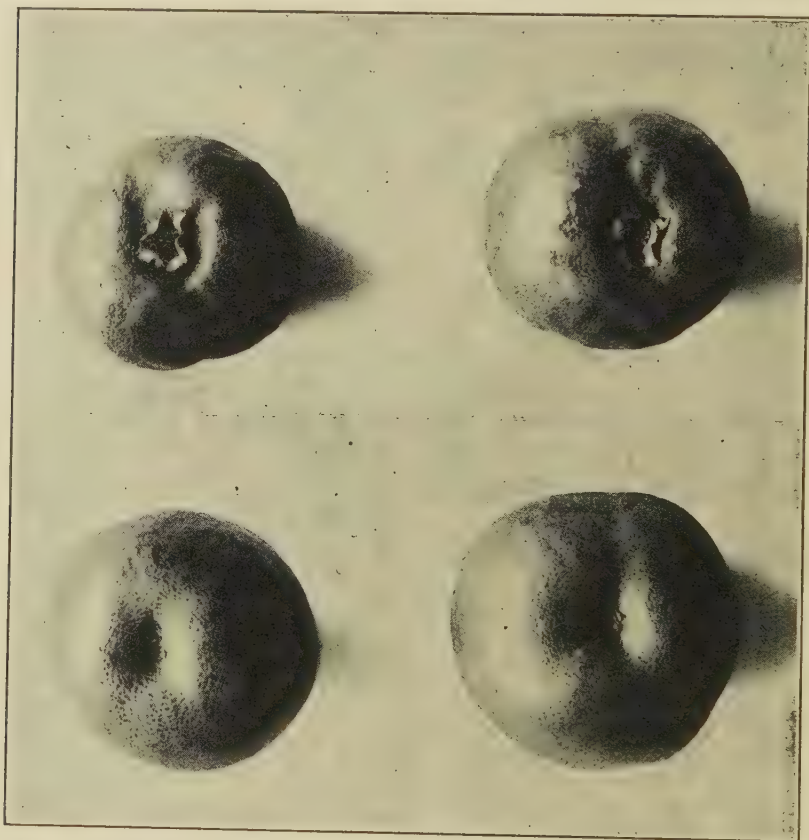


Fig. 1. Fruits of the Winter Nelis pear. The two upper are normal fruits with calyx attached. The two lower fruits show the result of finger nail emasculation which removed the calyx, yet there is practically no malformation.

breeding work, however, the writers feel that investigators should protect the emasculated and pollinated flowers with sacks to eliminate any possible chance of accidental pollination.

The writers have used the finger nail method of emasculation entirely, which consists of cutting through the calyx tube with the first or middle finger and thumb nails just below the point of attachment of the anthers, thus removing the calyx, corolla and anthers. This is the most rapid method and with a little practice 800 to 1,200 flowers can be emasculated per hour.

When tests were first started with the pome fruits, emasculation was performed with tweezers, removing petals and anthers. It was thought that the finger nail emasculation would seriously deform the fruit and might have an influence upon the results. By carefully checking results from pollinized flowers which had been emasculated with tweezers and by finger nails it was found that there was no difference in the sets obtained. There is practically no malformation of the fruits as is seen from the accompanying photograph. Therefore, since much more speed can be made by the finger nail method with no serious injury this method is now used entirely, not only with the drupes but also the pome fruits.

In the field tests it has not been at all uncommon to obtain "sets" in the artificial crosses of compatible varieties two to four times as great as the "normal set" which produced commercial yields.

Briefly summarized, the results obtained are as follows:

*Almonds.* All varieties of almonds tested have proved to be self-sterile. The list consists of twenty varieties. Two cases of intersterility have been found. Nonpareil and I.X.L. are intersterile, Languedoc and Texas are intersterile. All other varieties that bloom at the same time will pollinize each other though certain intercrosses give much higher "sets" than others.

*Apricots.* All commercial varieties of apricots grown in California are self-fertile and, therefore, do not require cross-pollination. In fact, from the results obtained the self-pollinations gave higher sets than did the crosses. Therefore, with the present day commercial varieties there is no pollination problem with apricots.

*Cherries.* Most serious difficulties have been encountered with sweet cherry pollination (*Prunus avium*). The greatest trouble encountered has been due to the question of nomenclature. In numerous cases apparently the same variety is known by different names. On the other hand in certain sections the same name is applied to different varieties. Again in certain cases, named varieties are probably groups of "strains" which vary in their pollination requirements. More than twenty varieties tested have proved self-sterile. The three most important commercial varieties; namely, Napoleon, Lambert and Bing, are inter-sterile. Several other cases of inter-sterility between unimportant commercial varieties have been found.

*Peaches.* Comparatively little work has been done with peaches as compared with other fruits. There is some belief that there is a pollination problem with the Tuscan Cling, and field observations indicate self-sterility of the J. H. Hale, though 1926 results at the University Farm do not indicate that the latter is self-sterile under California conditions.

*Pears.* With the exception of some of the very earliest blossoming varieties like Kieffer, LeConte and Alencon, all pear varieties overlap in their period of bloom. No cases of inter-sterility have been found to exist between pear varieties. Bartlett, the most important variety grown in California, is to a certain extent, self-fertile under interior valley conditions, but in most instances it is self-sterile in the Sierra Nevada Foothills. Pear varieties thus

far tested may be conveniently divided as self-sterile, self-fertile, and doubtful, as follows:

*Self-sterile.* Alencon, Bloodgood, B. S. Fox, Comet, Førelle, LeConte, Madeline and Winter Nelis.

*Self-fertile.* Comice, Flemish Beauty, Hardy and Howell.

*Doubtful.* Angoulême, Anjou, Bartlett, Bosc, Clapp Favorite, Clairgeau, Col. Wilder, Dana Hovey, Easter, Gifford, G. Morceau, Kieffer, P. Barry and Seckel.

*Plums and Prunes.* European Plums (*Prunus domestica*)—there is no inter-sterility among European plums. During some seasons there is considerable variation in blossoming season, and pollination of the late blossoming varieties with pollen from the earlier blossoming varieties is often difficult. European plums may be classed as self-sterile or self-fertile as follows:

SELF-STERILE		SELF-FERTILE	
Clyman	Washington	French	California Blue
Tragedy	Jefferson	Giant	Yellow Egg
Imperial	Quackenboss	Pond (Partially)	Coates 1418
President	Diamond	Grand Duke	(Double X)
Standard	Silver	(Partially)	Sugar
Robe de Sergeant			

Japanese and Japanese hybrid plums (*Prunus salicina*)—Most Japanese plums are self-sterile. Formosa and Gaviota are also inter-sterile. Tragedy will pollinize several Japanese varieties but is not pollinized by them. The following Japanese plums are self-sterile: Abundance, Apex, Burbank, Combination, Duarte, El Dorado, Formosa, Gaviota, Kelsey, Prize, Satsuma, Sultan, Upright and Wickson. Methley, Climax, Beauty and Santa Rosa are partially self-fertile but are greatly benefited by cross-pollination.

*Use of Bees.* The use of bees in orchards has proven to be very profitable, particularly with almonds, cherries, pears and plums. The usual procedure is to rent a number of colonies from commercial bee-keepers for the blossoming season. The bees are later removed. In the case of pears some trouble has been encountered following the use of bees because of spreading pear blight.

While a great mass of data has been collected, field tests are still being made. The most extensive work is being done with the cherry, hoping to isolate inter-fertile strains of our present commercial varieties and to test new varieties. With the other fruits mentioned above additional tests will be continued with new promising varieties.



# STUDIES ON THE STERILITY OF THE FRUIT TREES IN RUSSIA\*

W. PASHKEVITCH

*Institut de Botanique Appliquee, Leningrad*

As early as 1894 the success of fruit production by cross-pollination was pointed out by the Russian journalist Elpe (pseudonym) and from that time on the question was frequently discussed and brought up for consideration in general as well as in professional publications. (See *Novoje Vremja*, 1894 and 1898, Nos. 7915 and 7937; *Plodovodstvo* (Fruit Growing), 1895, p. 55; 1898, p. 100, 418, 534.) Waite in his paper published in 1895, "The Pollination of Pear Flowers," and in a second, 1898, "The Pollination of Pomaceous Fruits," directed the attention of botanists and fruit growers to the biological importance of the proper pollination and fertilization of the ovaries of fruit trees, a matter of importance for the yield of fruit trees in general and for the productiveness of certain varieties. (*Trans. Hort. Soc.*, London 5: 208-212, 1823.)

To be sure almost a century before, G. Swayne had already noted that since *Pyrus communis* is protogynous its flowers absolutely require pollination; but the minds of the scientists of that time were not sufficiently open to such biological questions and it was only toward the end of the 19th century that they were adequately appreciated. With Waite's paper the terms self-fertile and self-sterile were applied to the fruit crops in a way that struck a responsive chord in the minds of practical fruit growers.

## EXPERIMENTS BY PROFESSOR BELAJEV

In this manner the tests for the determination of the self-sterile and self-fertile varieties of fruit trees began not only in America but also in Europe. Such questions interested not only the western European scientists and fruit growers but also the Russian. Above all is here to be mentioned W. Belajev, Professor at the old University of Warsaw and Director of the Warsaw Pomological Garden. In 1899 he began self- and cross-pollination experiments on different pear and apple varieties in sufficient number and with methods adequate to present clearly and in detail the pollination problems.

The first of his series (*Plodovodstvo*, 1901, p. 204) concerns the question of the possibility of the self-pollination of blossoms. In these experiments whole inflorescences were enclosed in sacs of a closely woven cheese cloth and left to themselves. He obtained the following results:

---

\* This paper was presented to the Conference in English by Professor Madame J. Krasnasselsky-Maximow who also translated the manuscript from the Russian language to the German. The further translation as here printed was made by Dr. Illo Hein of the Department of Botany, Columbia University.

Pear Variety, Long Green.....	1	branch with	5	flowers gave	0	fruits
Apple Variety, Stepanovskoje.....	1	" "	4	" "	0	"
Apple Variety, Schirokoljitshiko.....	1	" "	8	" "	0	"
Apple Variety, Korobovka.....	1	" "	5	" "	0	"
Apple Variety, Ljimonnoje.....	1	" "	7	" "	0	"
Pear, <i>Pyrus ussuriensis</i> .....	1	" "	20	" "	0	"

On the basis of these results Prof. Belajev maintains that when all living organisms are excluded from the blossoms no fruit is formed on pear trees and apple trees.

The second series was concerned with pollination tests of pear trees and was designed to determine the effects of artificial pollination with the pollen of the same flower or with pollen from another flower of the same tree. The final result with the following varieties was:

Bartlett (Bon Chretien Williams).....	6	flowers gave	1	fruit
Doyenné du Comice.....	4	" "	0	"
Dean of Winter ("Doyenne d'hiver").....	3	" "	0	"
Early Duchess.....	1	" "	0	"
Beurré Capiaumont.....	5	" "	0	"
Seedless Bessemjanka—Russian.....	3	" "	0	"

For the third test series, pollen was taken from the same variety but from a different tree. In this case the following results were obtained:

	Flowers pollinated	Fruit obtained
Bartlett (Bon Chretien Williams).....	11	2
Passe Colmar.....	8	0
Beurré d'Amanlis.....	4	2
Beurré Diel.....	9	2
Doyenné du Comice.....	5	0
Early Duchess.....	5	4
Doyenné d'hiver.....	2	0
Beurré Capiaumont.....	5	2
Gansel's Bergamotte.....	5	0
Tonkovjetka (Russian Variety).....	1	0
Long Green.....	6	1

Finally in the fourth series cross-pollinations in ten combinations were carried out with the following results:

Seed parents	Pollen parents	Flowers	Fruits
Bartlett (Bon Chretien).....	× Gansel's Bergamotte.....	6	1
Bartlett (Bon Chretien).....	× Calabasse Tougard.....	6	3
Passe Colmar.....	× Calabasse Tougard.....	5	4
Passe Colmar.....	× Gansel's Bergamotte.....	4	2
Beurré Diel.....	× Gansel's Bergamotte.....	5	3
Doyenné d'hiver (Dean of Winter).....	× Tonkovjetka.....	2	2
Early Duchess.....	× Gansel's Bergamotte.....	3	1
Gansel's Bergamotte.....	× Calabasse Tougard.....	5	0
Tonkovjetka.....	× Doyenné du Comice.....	4	4
Tonkovjetka.....	× Beurré superfin.....	2	2

Through cross-pollination one apparently obtains the best results. Yet combinations occur here also which result in fruitlessness or only slight fruit production (Gansel's Bergamotte × Calabasse Tougard) (Bartlett × Gansel's Bergamotte).

"The problems resulting from our tests of the inter-relations of different varieties in pollination," Professor Belajev says, "must have definite practical

significance but for their complete elucidation numerous experiments will be necessary."

For apple trees Prof. Belajev made tests in similar series. Pollination of different apple varieties with pollen from the same tree gave the following results:

	Flowers pollinated	Fruits
Pear Apple Grooshevka.....	2	0
Duchovoje (Duft Apfel).....	5	0
Tchernoje Derevo (Black Tree).....	3	0
Zsarskij Schip (Kaiserdorn).....	4	0
Stepanowskoje .....Reine .....	5	0
Schirokoljitschiko .....	5	0
Kliujevskoje .....	3	2
Antonovka .....	4	1
Ljimonnoje (Zitronenapfel).....	4	2
Transparent White.....	4	0
*Borovinka (Oldenburg).....	4	0

Pollination with pollen of the same variety, but from a different tree gave:

Antonovka .....	4	flowers	gave	2	fruit
Borovinka .....	5	"	"	0	"
Baumann's Reinette.....	5	"	"	1	"

Pollination with pollen from a different variety gave:

Antonovka × Reine de Reinettes (Queen of Reinet) Winter Gold Pearmain.....	5	pollinated	flowers	gave	4	fruit
Antonovka × Linnaeus Pippin.....	1	"	"	"	1	"
Antonovka × Reinette de Canada (Golden Russet) .....	9	"	"	"	7	"
Tshornujie Derevo × Reine de Reinettes.....	3	"	"	"	0	"
Borovinka × Linnaeus Pippin.....	4	"	"	"	3	"
Red Astrakan × Linnaeus Pippin.....	3	"	"	"	2	"
Korobovka × Reinette de Canada.....	6	"	"	"	1	"
Transparent White × Reinette de Canada.....	2	"	"	"	0	"

The experiments with apple trees gave similar results to those with pears. Self-pollination is not always unfruitful. One also finds fruitlessness resulting from cross-pollination in some combinations but in general cross-pollination gives the best fruit production.

In addition to the above test series Prof. Belajev concerned himself with a series of other experiments involving the pollination of various pear and even apple varieties with pollen from *Pyrus ussuriensis*. The following results were obtained:

<i>Pyrus ussuriensis</i> × Doyenné du Comice.....	6	pollinated	flowers	gave	4	fruit
<i>Pyrus ussuriensis</i> × Beurré Diel.....	5	"	"	"	1	"
<i>Pyrus ussuriensis</i> × Beurré d'Amanlis.....	5	"	"	"	4	"
<i>Pyrus ussuriensis</i> × Calebasse monstre.....	7	"	"	"	5	"

By pollination of other pear trees with pollen of *Pyrus ussuriensis* the following results were obtained:

\* The variety Borovinka and the Duchess of Oldenburg are considered identical.

Doyenné d'hiver × <i>Pyrus ussuriensis</i> .....	4	flowers	gave	2	fruits
Early Duchess × <i>Pyrus ussuriensis</i> .....	2	"	"	1	"
Beurré Capiaumont × <i>Pyrus ussuriensis</i> .....	1	"	"	3	"

The pollination series consisting of flowers of *Pyrus ussuriensis* with the pollen of apple varieties, Canadian Reinette and Baumann's Reinette were unusually interesting. Seven pollinated flowers of *Pyrus ussuriensis* gave 4 fruits.

Out of these pollination tests with *Pyrus ussuriensis* the following may be learned: First, that a distant relationship between the pollinated forms gives more productive combinations; second, that we have in the cases given examples of successful crossings, not only with forms of different species (*Pyrus ussuriensis* and *Pyrus communis* or others) but also with different genera (*Pyrus* and *Malus*). Unfortunately it must be accepted that the fruits of *Pyrus ussuriensis*, obtained by crossing it with the two above-mentioned apple varieties possessed no seed capable of germination, for Prof. Belajev states that the fruits produced by crossing pears with *Pyrus ussuriensis*, have given viable seeds in both directions and that seedling trees have thrived in the Warsaw garden, as opposed to this he does not mention the trees or the seeds which he had obtained from the crossing of *Pyrus ussuriensis* with apples.

Out of the tests with *Pyrus ussuriensis* it follows that they are to be looked upon as capable pollinators for pears and their presence in pear orchards is therefore very desirable. Seed can be obtained from their fruits and these again can be used for the planting of their wild stock.

For this reason the former Department of Agriculture gave appropriate attention to *Pyrus ussuriensis* and ordered seeds of this pear from Japan for purposes of propagation and cared for the necessary extended multiplication for experimental purposes in experimental plots.

### EXPERIMENTS BY P. MOSKALJEV

P. Moskaljev tested the pollination requirements of certain fruits in his garden in Tschudnowo, Gouv. Wolhynien, according to the statements of Prof. Belajev (*Plodovodstvo*, 1902, p. 597). Although he was unable to notice any self-sterility in the pears Clairgeau, Regentin, and Comtesse de Paris he could ascertain that the fruits obtained by self-pollination contained only a few poorly developed seeds that would not germinate. Thus two selfed fruits of the pear Clairgeau contained only four rudimentary seeds, one selfed fruit of Regentin contained two seeds and one fruit of Comtesse de Paris contained three seeds.

Against this he obtained through pollination with the variety Dean of Winter (Doyenné d'Hiver) 13 seed kernels from two fruits; through pollination with the variety Beurré d'Hardenpont out of 1 fruit 9 seed kernels, and with pollination by Comtesse de Paris out of 2 fruits 20 seed kernels. All these seed kernels were in these cases perfectly developed.



## COLLECTIVE EXPERIMENTS OF 1911-1914

In the years 1911 and 1912 the author of this paper undertook an experimental study of the results of self- and cross-pollination in fruit species. These experiments were performed in the Pensa Horticultural School, in the village of Gorutjishki, Gouv. Minsk in the neighborhood of Koydanowo by W. Pashkevitsch, in the Nikita Botanical Garden by Professor W. Ljubimenko and in the territory of the former Kowno government, in the estate Bersheniki, Kreis Alexandrovsk by Professor A. Hrebznizki.

## EXPERIMENTS IN THE PENSA HORTICULTURAL SCHOOL

The tests in the Pensa Horticultural School were carried on by simply enclosing the twigs with cheesecloth before the beginning of flowering, without the application of artificial pollination with their own or of pollen of the same variety. Cross-pollination tests were also made in another series. The results obtained were as follows:

## RESULTS OF SELF-POLLINATION

Varieties	Spurs and blossoms enclosed		Fruit Settings	Fruits Harvested	%
	Spurs	Flowers			
Antonovka, 1 main branch, 1911.....	..	..	9	9	?
Antonovka, 1 twig, 1911.....	24	78	8	8	10.25
Antonovka, entire tree, 1912.....	8	35	10	0	0
Antonovka, twig, 1912.....	8	35	2	1	2.9
Borovinka, $\frac{1}{2}$ of tree, 1911.....	?	?	3	0	0
Borovinka, twig, 1911 .....	20	101	0	0	0
Borovinka, 5 twigs .....	?	?	0	0	0
Borovinka, No. 4, 1912.....	..	..	0	0	0
Borovinka, No. 3, 1912.....	4	17	2	2	12
Gray Anis, twig.....	5	33	1	1	3
Gray Anis, control twig.....	2	13	7	5	38.5
Granat, twig .....	11	75	6	3	4
Granat, control twig.....	4	25	10	2	8
Skrizshapel .....	?	33	1	0	0
Skrizshapel, control twig.....	3	16	8	2	12.5

## RESULTS OF CROSS-POLLINATION

Varieties	Spurs and blossoms enclosed		Fruit Settings	Fruits Harvested	%
	Spurs	Flowers			
Anis Gray $\times$ Antonovka .....	3	22	6	4	18
Anis Gray $\times$ Borovinka .....	1	6	2	1	16.5
Anis Gray $\times$ Granat .....	1	6	2	2	33
Borovinka $\times$ Antonovka .....	1	4	1	1	25
Borovinka $\times$ Anis .....	4	14	11	3	25
Borovinka $\times$ Granat .....	1	6	6	3	50
Granat $\times$ Antonovka .....	2	10	2	1	10
Granat $\times$ Anis .....	3	17	7	3	20
Skrizshapel $\times$ Antonovka.....	3	10	4	3	30
Antonovka $\times$ Anis .....	4	14	4		Fruit buds
Antonovka $\times$ Amtmann's Apfel .....	3	13	2		
Antonovka $\times$ Titovka .....	4	16	4		torn off
Antonovka $\times$ Skrut .....	4	12	4		by wind

Self-pollination tests carried on in the above manner at Gorutjschiski in the Government Minsk by the author gave the following results:

	Number flowers enclosed	Fruit Settings	Fruit Harvested	%
Ostheimer Weichsel, entire tree, in 1911.....	166	99	..	..
Ostheimer Weichsel, 4 young trees.....	..	31	0	..
Osteheimer Weichsel, control.....	..	60	3	..
Autumn Bergamotte.....	85	0	0	0
Pear, Iljinka.....	101	2	2	2
White Summer Sugar Apple, Branch.....	1000	0	0	*
Antonovka .....	200	0	0	*
Boiken .....	75	0	0	*
Morkvjanka Pear .....	1200	0	0	*
Longfield .....	505	7	7	1.38
Titovka .....	300	5	5	1.67**
Borovinka, 2 branches.....	465	0	0	..
Transparent White, 1 branch.....	50	0	0	..
Korobovka, 2 branches.....	180	0	0	..
Antonovka, 1 twig.....	15	0	0	..
Amtmannsapfel, 1 branch.....	450	0	0	..
Alexander, 2 twigs.....	50	0	0	..
Tchernogoos, 2 twigs.....	46	0	0	..
Boiken, 5 twigs.....	75	0	0	..
<i>Malus baccata</i> , on two trees.....	65	0	0	..
<i>Prunus Pensylvanica</i> .....	585	0	0	***
<i>Prunus Pensylvanica</i> , control shrub.....	?	0	0	***
<i>Amelanchier ovalis</i> .....	540	..	..	**
<i>Amelanchier ovalis</i> , control.....	?	..	..	**

#### EXPERIMENTS AT LUGA, GOVERNMENT LENINGRAD

We shall add to this still another experiment which was executed according to our plan in the year 1914 in the state Nikolajewskoje with the apple variety Antonovka\*. Instead of merely enveloping a twig in cheese cloth in this experiment, an entire young Antonovka tree and its crown was enclosed on four sides by a framework upon which a closely woven cheese-cloth was stretched, the whole tree being covered from top to bottom and all openings carefully closed up. This enclosure protected the tree from injury and could not have exercised any influence (without the intervention of insects) on normal self-pollination. The fallen fruit settings were counted daily and as a result it was found that the three had thrown off a total of 1198 fruit settings (between June 14th and August 14th, 516 large and small fruit settings and 682 injured). Only 12 large apples and one medium-sized apple were harvested and one large apple which had fallen before the readings were taken. In this manner about 1.2 per cent completely developed apples were obtained but of these four were entirely seedless, two contained only one weakly developed seed kernel, six had a single fully developed seed kernel and only one was filled with completely developed seeds.

This completely demonstrated the inability of fruit production of Antonovka by self-pollination, since 1.2 per cent undeveloped fruit is to be considered negligible if not entirely fruitless.

\* Good yield on the rest of the tree.

\*\* Rich harvest.

\*\*\* No fruits on entire tree.

\*Nautshnoje Plodovodstvo, Scientific Fruit Growing, 1915, 2nd annual set, 2nd volume published by the Russian Fruit Growing Society.

Cross-pollinations carried in Gorutjischki Government Minsk gave the following results:

Seed	Pollen Parent	Flowers enclosed and pollinated	Fruit July 5	Fruit Sept. 17	%
Antonovka	× White Summer Sugar Apple	5	3	3	60
Antonovka	× White Summer Sugar Apple	11	6	5	48
Antonovka	× Longfield	7	3	3	46
Antonovka	× Longfield	6	3	2	33
Boiken	× Red Stettiner	27	0	0	..
Boiken	× Kartoffljanka Potato-apple	10	4	4	48
Boiken	× Yellow Sweet	32	10	8	25
Transparent White	× Longfield	46	5	5	41.7
Transparent White	× White Summer Sugar Apple	24	3	3	12.5

In the year 1912:					
Transparent White	× Antonovka	8	0	0	..
Transparent White	× Borovinka	8	0	0	..
Borovinka	× Grosse Kasseler Reinette	20	0	0	..
Borovinka	× Alexander	15	0	0	.. *
Longfield	× Transparent White	10	9	..	90 *
Longfield	× Grosse Kasseler Reinette	12	7	..	58
Amtmannsapfel	× Borovinka	50	0	0	.. **
Alexander	× Borovinka	4	1	..	25
Small Sweet	× Longfield	25	0	0	.. *
Antonovka	× Transparent White	16	0	0	.. **
Tschernogoos	× Borovinka	10	1	1	10 **

As a result of the general small garden harvest the results of the year 1912 were less decisive than those obtained in the year 1911.

#### EXPERIMENTS CONDUCTED AT NIKITA BOTANICAL GARDEN (KRIM) BY PROFESSOR H. LJUBIMENKO

Prof. H. Ljubimenko set up in the Nikita Botanical Garden crossing experiments with 2 peach and 7 apple varieties, in order to investigate mainly the effect of pollen of other species on change in the quality and peculiarities of the fruit.

In this manner the pollination of peaches yielded the following:

Baron Dufur	× De Brussa, 40 pollinated flowers yielded	12 ripe fruits
De Brussa	× Baron Dufur, 40 pollinated flowers yielded	12 ripe fruits

For the apple varieties the following data were obtained:

Winter Gold Pearmain	× Baumann's Reinette	18 ripe apples
Winter Gold Pearmain	control	77 ripe apples
Baumann's Reinette	× Golden Russet	26 ripe apples
Baumann's Reinette	control	14 ripe apples
Winter Gold Pearmain	× Pineapple Reinette	8 ripe apples
Calville Red Autumn	× White Calville of Winter	7 ripe apples
Candil Ssinap	× Crede's Pigeon	few fruits

These trials demonstrated conclusively the absence of sterility in the above quoted crossing combinations. Unfortunately since the number of flowers which were pollinated is not known, there is not presented any possibility of an opinion concerning the percentage which falls to each successful combination.

\* Good yield from the tree.

\*\* Very poor yield on the tree.

# EXPERIMENTS BY PROFESSOR A. S. HREBNJITZKY, GOVERNMENT KOWNO

The pollination tests performed by Professor A. S. Hrebnjitzky of the former Government Kowno led to the following results:

		Number of Pollinated Flowers	Fruit on May 18	Settings on June 14	Fruit Harvested Aug. 23	%
Antonovka	× Moscow Pear Apple.....	6	6	4	4	66.67
Antonovka	× <i>Malus baccata</i> (Kitaika)...	11	11	8	6	54.54
Antonovka	× Transparent White .....	7	7	6	5	71.43
Antonovka	× Littauer Sugar Apple.....	9	9	6	6	66.67
Antonovka	× Witjebesk Putjimka .....	6	6	0	0	0
Antonovka	× Berzsheniky Pineapple.....	20	20	2	2	10
Antonovka	× Longfield .....	17	17	1	1	5.88
Antonovka	× Amtmannsapfel .....	15	15	0	0	0
Antonovka	× Papirovska .....	15	15	3	1	6.67
Antonovka	× Antonovka, artificially pol- linated .....	31	31	0	0	0
Antonovka	× Antonovka, self-pollinated in bags .....			0	0	0
Antonovka	× Antonovka, self-pollinated in bags .....	13	13	0	0	0
Antonovka	× scars cut off from partheno- carpic test castrated.....	229		18	15	6.5
Berzsheniky	Pineapple × Amtmann's Apple .....	20	16	2	0	0 *
Berzsheniky	Pineapple × Littauer Sugar Apple .....	18	12	0	0	0 *
Berzsheniky	Pineapple × Longfield.....	9	0	0	0	0 *
Berzsheniky	Pineapple × Papirovska.....	12	0	0	0	0 *
Berzsheniky	Pineapple × Alexander.....	11	0	0	0	0 *
Berzsheniky	Pineapple × Antonovka.....	14	0	0	0	0 *
Berzsheniky	Pineapple × Berzsheniky Pineapple .....	10	0	0	0	0 *
Berzsheniky	Pineapple × Witjebesk Put- jimka .....	7	0	0	0	0 *
Berzsheniky	Pineapple × Harbert's Rei- nette.....	10	0	0	0	0 *
Berzsheniky	Pineapple × <i>Malus baccata</i> ..	37	32	0	0	0 *
Berzsheniky	Pineapple × Red Striped....	11	10	0	0	0 *
Berzsheniky	Pineapple × Purple Red Anis	10	9	4	4	40 *
Alexander	× Papirovska .....	17	10	5	1	6
Alexander	× Antonovka .....	25	7	0	0	0
Alexander	× Littauer Sugar Apple.....	12	1	1	1	9
Alexander	× Amtmann's Apple.....	10	3	1	1	10
Alexander	× Harbert's Reinette (old tree)	10	4	3	2	20
Alexander	× Alexander .....	7	0	0	0	0
Alexander	× Berzsheniky Pineapple (old tree) .....	10	7	4	2	20
Alexander	× Berzsheniky Pineapple (young tree).....	11	3	1	1	9
Papirovska	× Antonovka.....	22	20	13		40
Papirovska	control tree.....	11	11			99.9
Papirovska	control tree with girdling.....	4	4			100

These experiments show conspicuously the sterility of the variety Berzsheniky Pineapple in all combinations with the exception of the combination Berzsheniky Pineapple × Anis purple red. Professor Hrebnjitzky explains this by the fact that in the year concerned (1912) all pineapple trees in his garden were fruitless and that the general productive capacity of the tree had also made a general impression on the few artificially pollinated blossoms which succeeded in unfolding. This fact should be kept in mind, in



order that the experimenters may not be misled to false conclusions by one or the other crossing combinations. Even more convincing than the Berzheniky Pineapple has shown in the above case, the capacity of the pollen of the Purple red Anis to be used with good success for pollination is revealed.

#### NEW TEST SERIES IN 1924

The events of the world war and revolution which entered after the preparation and publication of these papers entirely prevented any further work in the field of pollination experiments. Not until the year 1924 did the author succeed when commissioned by the horticultural section of the Institute of Experimental Agronomy and later when commissioned by the All-Russian Institute for Applied Botany and New Cultures, in organizing new comprehensive experiments concerning self- and cross-pollination of fruit varieties. The summarizing of the work appeared in 1924 in the Bulletin of Applied Botany (Vol. XIV, No. 3, 1924-1925). Out of this volume the following data may be gathered:

		Place of Ex- periments*	Fruit Settings			%
			Pollinated Flowers	after two weeks	Ripe Fruits	
Antonovka	× Antonovka	D.S.	75	4	0	0
Antonovka	× Antonovka	Ch.G.	58	14	1	1.7
Antonovka	× Borovinka	Ch.G.	100	7	1	2.4
Antonovka	× Transparent White	D.S.	37	9	1	2.7
Antonovka	× Titovka	Ch.G.	64	14	3	4.7
Antonovka	× Longfield	Ch.G.	70	14	4	5.9
Antonovka	× Barlovskoje	D.S.	16	2	0	0
Borovinka	× Borovinka (June 3)	Ch.G.	45	6	2	4.4
Borovinka	× Borovinka (July 3)	Ch.G.	12	1	0	0
Borovinka	× Alexander	Ch.G.	34		10	29.4
Borovinka	× Titovka	Ch.G.	64	0	6	9.3
Borovinka	× Longfield	Ch.G.	18		2	11.1
Titovka	× Titovka	Ch.G.	51	13	0	0
Titovka	× Titovka (Dsek)**	D.S.	100	89	63	63
Titovka	× Titovka (Jegor)***	D.S.	25	1	0	0
Titovka	× Antonovka	D.S.	115	82	44	34.8
Titovka	× Borovinka	Ch.G.	40	12	5	12.5
Titovka	× Longfield	Ch.G.	55	17	4	7.2
Titovka	× White Astrakan	D.S.	25	14	?	?
Titovka	× Transparent White	D.S.	50	30	16	32
White Astrakan	× Transparent White	D.S.	50	29	?	?
White Astrakan	× Transparent White	D.S.	33	6	6	18
Transparent White	× White Astrakan	D.S.	22	2	1	4
Transparent White	× White Astrakan	D.S.	3	0	0	0
Transparent White	× Antonovka	D.S.	64	18	9	14.06
Transparent White	× Transparent White	D.S.	49	17	17	34
Transparent White	× Barlovskoje	D.S.	87	24	?	?
White Ribbed	× Titovka	D.S.	13	6	5	44
Alexander	× Alexander	Ch.G.	100	68	1	3.1
Babushkino	× Wealthy	W.	100	2	0	0
Wealthy	× Babushkino	W.	100	29	25	25
Landsberg Reinette	× Landsberg Reinette	Ch.G.	19	6	0	0
Skljanka Kursk	× Skljanka Kursk	Ch.G.	52	19	0	0
Skvosnoi Naliv	× Titovka	D.S.	12	4	4	33
Tschernogoos	× Titovka	D.S.	23	18	6	24

\* The abbreviations used under "Place of Experiments" are to be read: D.S. is Djetskoe Selo, garden of the Agricultural Institute in Leningrad. Ch.G. is Horticultural Division of the Experiment Station in Charkow. W. is Agricultural Institute at Woronesch.

\*\* Experimenter, Miss Dsenonitzkajia.

\*\*\* Experimenter, Miss Segoriva.

## EXPERIMENTS AT THE MYSSOVSKAJA STATION

At the same time and in the same year, 1924, crossing experiments were carried on at the Myssovskaja Experiment Station for fruit and vegetable growing, of the Moscow Agricultural Society by A. Petroff, Assistant in the Timerjaseff Agricultural Academy. His tests resulted in the following combinations:

Common Antonovka	×	Amtmann's Apple	.....	=	51.4%	ripe fruits
Common Antonovka	×	Zimmt Striped	.....	=	42.8%	" "
Common Antonovka	×	Borovinka	.....	=	37.7%	" "
Common Antonovka	×	Papirovka	.....	=	33.3%	" "
Common Antonovka	×	Anis Striped	.....	=	33.3%	" "
Common Antonovka	×	Arkad, long yellow	.....	=	28.5%	" "
Common Antonovka	×	Common Antonovka	.....	=	0	
Borovinka	×	Pear Apple, large	.....	=	50 %	" "
Borovinka	×	Arkad, long yellow	.....	=	42.5%	" "
Borovinka	×	Kitaika Sanin	.....	=	42.1%	" "
Borovinka	×	Antonovka, yellow	.....			
		Karobkower Garden near Moscow	.....	=	35.9%	" "
Borovinka	×	Common Antonovka	.....	=	34.2%	" "
Borovinka	×	Zimmt Striped	.....	=	18.2%	" "
Borovinka	×	Borovinka	.....	=	0	
Borovinka		isolated from insects	.....	=	0	
Pear Apple, large	×	Pear Apple Moscow	.....	=	45.9%	" "
Pear Apple, large	×	Pear Apple, large	.....	=	0	
Pear Apple, large	×	Zimmt Striped	.....	=	44.4%	" "
Pear Apple, large	×	Antonovka	.....	=	36.8%	" "
Pear Apple, large	×	Borovinka	.....	=	33.3%	" "
Pear Apple		control tree	.....	=	41 %	" "
Zimmt Apple, smoky colored	×	Antonovka	.....	=	20 %	" "
Zimmt Apple, smoky colored	×	Borovinka	.....	=	17.4%	" "
Zimmt Apple, smoky colored	×	Zimmt Striped	.....	=	2 %	" "
Zimmt Apple, smoky colored,		control tree	.....	=	33 %	" "

## EXPERIMENTS AT THE MYSSOVSKAJA EXPERIMENT STATION IN THE YEAR 1925

In 1925 self- and cross-pollination experiments were continued in the Myssovskaja Experiment Station as well as in the neighborhood of Leningrad in Djetskoje Sselo and in the Horticultural School at Krasnaja Slavjanka. We are in a position here to give only a short summary of the work carried on in

	Flowers Pollinated	Fruit Settings		Fruit Harvested		%
		May 27	June 22	Aug. 8		
Papirovka	×	Arkad, long yellow	22	12	10	45.5
Papirovka	×	Borovinka	18	7	2	11.1
Papirovka	×	Kitaika Sanin	16	6	2	12.5
Papirovka	×	Kitaika Sanin, yellow	26	9	9	34.6
Papirovka	×	Papirovka	24	20/83.3%	0	0
Papirovka, only two flowers and entire flower stand isolated	12-27	0	0	0	0	0
Fall Striped	×	Antonovka	16	9	8	12.5
Fall Striped	×	Wealthy	22	8	6	9.09
Fall Striped	×	Korobovka	20	4	4	10
Fall Striped	×	Zimmt Striped	20	4	0	0
Fall Striped	×	Amtmann's Apple	18	5	3	0
Fall Striped, flowers isolated	13	5	4	1	7.7	
Borovinka	×	Arkad, long yellow	22	5-18z. June 5	10 s'dl's Sept 10	45.45
Borovinka	×	Kitaika Sanin	17	16	" 1 "	5.9
Borovinka	×	Antonovka	24	20	" 11 "	45.8
Borovinka	×	Zimmt Striped	18	8	" 9 "	50
Borovinka	×	Borovinka	16	13	2 fruit seedless	12.5
Borovinka, flowers isolated	43	24	.. 2	" "	" "	4.65

this Station. Here the effect of the corresponding pollen of its own and of the pollen of other varieties were tested out only on the Papirovka, Amtmann's Apple and Borovinka trees. The results may be gathered from the list.

### EXPERIMENTS IN THE HORTICULTURAL SCHOOL AT KRASNAJA SLAVJANKA

In the Horticultural School at Krasnaja Slavjanka in the neighborhood of Djetskoje Sselo, Government Leningrad, cross-pollination as well as self-pollination experiments were made with apple varieties. For self-pollination experiments twigs with blossoms were, as usual, enclosed in isolation sacs and the unfolding blossoms were artificially pollinated with pollen of the same variety. Indeed for all the varieties 50 blossoms were chosen (only two to a stand), the remaining ones were cut off. The following varieties showed themselves to be completely self-sterile: Reinette (Golden); Skrizshapfel; Petersburg Rose Apple, Borovinka, Common Transparent White, Velvet-Anis, Petersburg Plodowitka, Clara Apple, White Ribbed, Arabka, Serinka, Virginia Rose Apple, Revaler Pear Apple, Alexander, Red Eiserapfel, Zolotarevka, Red Sugar Apple, Kramers Glass Apple, Red Wine Apple, Sommer-Borsdorfer and Kegelformiger Borsdorfer.

Only a few varieties possessed, if not to a very high degree, the capacity for self-fertility. The highest productive percentage, 34, was attained by Raspberry Apple from Livland; Suislepper gave 10%, White Astrakan 6%, Anis 5%, Amtmannsapple 4%, and Boiken, Zimmt-Striped Thorough Transparent (Skvosnoi Naliv), German Skrut, Poloomiron, Cream Apple—each 2 per cent.

	First Observation	Second Observation	Fruit Yield
Alexander × Titovka.....	92%	0%	0
Alexander × Borovinka .....	80	6	0
Alexander × Antonovka .....	82	0	0
Alexander × Boiken .....	66	0	0
Alexander × Red Eiser .....	32	8	3
Alexander × Cream Apple .....	80	18	3
Alexander × Zimmt Striped .....	68	8	4
Alexander × Livland Raspberry .....	62	14	1
Alexander × Thorough Transparent.....	46	18	4
Alexander × White Astrakan .....	86	12	2
Alexander × Amtmann's Apple .....	68	8	0
Alexander × Reval Pear Apple .....	76	10	1
Alexander × Transparent White .....	84	20	0
Antonovka × Wine Apple .....	49	9	3
Antonovka × Titovka .....	46	11	5
Antonovka × Alexander .....	76	38	6
Antonovka × Borovinka .....	68	35	8
Kegelform. Borsdorfer × Antonovka.....	14	12	1
Red Eiser × Alexander.....	38	10	1
Reval. Pear Apple × Antonovka .....	70	28	0
Reval. Pear Apple × Alexander .....	88	22	4
White Astrakan × Alexander.....	82	36	7

For crossing experiments 50 blossoms, two out of each stand, were taken in all combinations and only for Antonovka 100 blossoms were taken. The pollination took place between the first and sixth of June. The first observa-

tion, between June 24th and June 27th, showed a high percentage of fruit settings (32 to 92 per cent), yet the second observation, between July 8th and July 15th showed some combinations at zero and in others the percentage of the remaining fruits had fallen considerably, as will be seen from the above tabulation.

### TESTS IN DJETSKOJE SSELO

In the year 1925 self-pollination tests were carried on with apple varieties in Djetskoje Sselo in the Garden of the Leningrad Agricultural Institute, yielding the following results:

	Blossoms	Fruit settings after 2 weeks	Fruit harvested	%
Antonovka pollinated with its own pollen.....	101	24	9	9
Antonovka wrapped in cheesecloth and unpol- linated .....	111	16	1	.9
Antonovka control twig.....	106	32	21	20.8
Transparent White, self-pollinated.....	110	0	0	0
Transparent White, wrapped in cheesecloth...	279	0	0	0
Transparent White, control branch .....	114	58	45	39.5
Alexander, self-pollinated.....	125	..	0	0
Alexander, enclosed .....	139	0	0	0
Alexander, control twig .....	88	17	7	8.2
Tshornogus, self-pollinated .....	43	6	1	2.4
Tshornogus, enclosed .....	160	13	1	0.6
Tshornogus, control twig .....	108	11	0	0
Transparent Yellow, enclosed .....	85	2	0	0
Transparent Yellow, control twig .....	106	8	5	4.7
Borovinka, enclosed .....	87	0	0	0
Pypka Ljubskaja, enclosed twig.....	50	0	0	0
Pypka Ljubskaja, control twig.....	38	3	3	8.4
Pypka Ljubskaja, entire tree in cheesecloth...	221	31	0	0
White Apple, pollinated.....	10	0	0	0
White Apple, in cheesecloth.....	2	0	0	0

The figures in the last two tables speak for themselves; yet before we draw any final conclusions from the above quoted materials we should like to present the report which came to us from Prof. J. Kononov concerning the self- and cross-pollination tests with the varieties Kursk Golden Reinette, Fameuse, and Haas in his gardens at the Voronesh Horticultural Institute. The trees of these varieties were young, 16-year-olds. The following data have been received:

	Flowers pollinated	Fruit settings	Fruit harvested	%
Fameuse × Haas .....	100	81		
Fameuse × Fameuse .....	50	z. 22. V	33	33
Fameuse tests of parthenocarpie.....	54	0	0	0
Haas × Fameuse .....	100	24	0	0
Haas × Haas .....	50	31	14	14
Haas × Kursk Golden Reinette .....	50	2	0	0
Haas tests of parthenocarpie.....	100	34	11	11
Kursk Golden Reinette × Haas.....	50	0	0	0
Kursk Golden Reinette × Kursk Golden Reinette	100	96	52	52
Kursk Golden Reinette tests of parthenocarp...	50	0	0	0
	50	0	0	0

During these experiments a considerable number of fruits fell from the trees as a result of injury by the fungus *Monilia fructigena* and the fruit



destroyer, *Carpocapsa pomonana*, and on the other hand a part of the fruit settings were torn down by wind and strong rainfalls.

The multiplicity of the natural conditions in U. S. S. R. has also produced a great multiplicity of fruits which are distributed throughout this country. Besides their own Russian varieties, numerous foreign (including American) varieties are cultivated. There exists, according to this, a vast work in the field of investigation of reciprocal relations of pollination processes between all varieties that may be cultivated here. Although such investigations, which until now have been carried on in America as well as in western Europe offer us a wealth of finished material, nevertheless, we are taught that a change in the growth conditions of the corresponding native environment and perhaps even of culture conditions themselves, other results may be achieved. It is just on this account that we must carry out investigations in this country, in order to clarify and establish the results as completely as possible.

### SUMMARY

On the basis of the none too numerous experiments so far conducted, the following conclusions may be drawn:

1. According to the experiments conducted by Prof. W. Belajev, the pollination with the pollen of the same flower or flowers from the same tree, or even with the pollen from flowers of other trees belonging to the same variety, gave in all classes entirely the same results.

2. Similar experiments showed that *Pyrus ussuriensis* may be regarded as a good pollinator for pear varieties which originated from other forms.

3. *Pyrus ussuriensis* is even capable of successfully pollinating several apple varieties as, for example, Canadian Reinette and Baumann's Reinette, yet no seed capable of germinating can be obtained through this pollination. Even *Pyrus ussuriensis* itself produces no fruit through self-pollination.

4. The experimental data of Prof. Belajev concerning self-sterility of the apple varieties Stepanovskoje, Schirokoljitschiko, Duftapel, Black Tree, Kaiserdorn, Transparent White, Korobovka, Lemon Apple, Borovinka, as well as of the pear varieties Long Green, Bartlett, Doyenne du Comice, Dean of Winter, Seedless, Passe Colmar, Gansel's Bergamotte and Tonkovjetka are only of conditional importance, because the pollinations were carried on only with a small quantity. The self-sterility of *Pyrus ussuriensis* can, however, be unquestionably recognized.

5. On the basis of other experiments the following apple varieties are to be considered self-sterile: Antonovka (partly), Borovinka (greater part), Skrizshapfel, White Summer Sugar Apple, Boiken, Transparent White, Korobovka, Amtmann's Apple, Alexander, Tchorogooos, *Malus baccato* (?), Titovka, Landsberger Reinette, Kursk Glass Apple, Large Pear Apple, Papirovka, Kursk Golden Reinette, Velvet Anis, Petersburg Rose Apple, Petersburg Plodovitka, Transparent White, White Ribbed, Arabka, Serinka, Virginia Rose Apple, Revel Pear Apple, Red Eiser, Solotarjovka, Red Sugar Apple, Kramer's Glass Apple, Red Wine Apple, Summer Borsdorfer, Conical

Borsdorfer, Transparent White, Skvosnoi, Transparent Yellow, Ljubskaja, Pipka, White, Fameuse, and Haas.

6. Sterility as a result of cross-pollination has manifested itself in the following combinations:

(a) Prof. Belajev.—Gansel's Bergamotte  $\times$  Calebasse Tougard, and Black Tree  $\times$  Winter Pearmain.

(b) In Gorutjischki, White Russia, Gouv. Minsk.—Boiken  $\times$  Red Stettin, Transparent White  $\times$  Borovinka, Borovinka  $\times$  Alexander, Amtmann's Apple  $\times$  Borovinka, and Small Sweet  $\times$  Longfield.

(c) In the tests by Hrebritzky.—Antonovka  $\times$  Putjimka Witebskaja, Antonovka  $\times$  Amtmann's Apple, Antonovka  $\times$  Harbert's Reinette, Alexander  $\times$  Antonovka, and Berzsheniky Pineapple in all combinations for the purpose of cross-pollination with the exception of that of the Purple Red Anis.

(d) Tests in Djetskoje Sselo.—Antonovka  $\times$  Barlovskoje.

(e) Tests at the Myssovskaja Experiment Station.—Amtmann's Apple  $\times$  Zimmt Striped.

(f) Tests in the Krasnaja Slavjanka Horticultural School.—Alexander  $\times$  Titovka, Alexander  $\times$  Borovinka, Alexander  $\times$  Antonovka, Alexander  $\times$  Boiken, Alexander  $\times$  Amtmann's Apple, Alexander  $\times$  Transparent White, Revel Pear Apple  $\times$  Antonovka.

7. The highest percentage (50-100) by cross-pollination of actually ripened fruit was obtained by Prof. Belajev in Warsaw in a series of combinations and a fairly large per cent (40-71) in just a few.

The percentage in the experiments of Prof. A. Hrebizky was 40 for Berzsheniky Pineapple  $\times$  Purple Red Anis, 66 for Antonovka  $\times$  Moscow Pear Apple, 54.5 for Antonovka  $\times$  *Malus baccata*, 71.4 for Antonovka  $\times$  Transparent White; 66.7 for Antonovka  $\times$  Littauer Sugar Apple and 40 for Papirovka  $\times$  Antonovka.

In the experiments at the Myssovskaja Experiment Station in 1924, the percentage of all, with the exception of the combinations destined for cross-pollination, were 28.5 to 51.4 and attained 18.2 only in the combination Borovinka  $\times$  Zimmt Striped, and from 2 to 20 per cent in combinations of Smoky Grey Zimmt apfel with three other varieties. As against this in 1925 the per cent of success varied in only 5 combinations between 34 and 50. These combinations were: Papirovka  $\times$  Arkad long yellow, 45.5 per cent; Papirovka  $\times$  *Prunus prunifolia* variety, 34.6 per cent; Borovinka  $\times$  Arkad Long Yellow, 45.5 per cent; Borovinka  $\times$  Zimmt Striped, 50 per cent. In the remaining cases it did not exceed 12.5 per cent.

In the experiments in Djetskoje Sselo in the year 1924, the percentage of success of the different combinations did not exceed 44, for Borovinka  $\times$  Alexander gave 29.4 per cent; Titovka  $\times$  Antonovka, 34.8 per cent; Titovka  $\times$  Transparent White, 32 per cent, and White Ribbed  $\times$  Titovka, 44 per cent.

The experiments at the Agricultural Institute at Woronesch showed the

following percentages: Wealthy  $\times$  Babuschkino, 25; Kursk Golden Reinette  $\times$  Haas, 52, and Fameuse  $\times$  Haas, 33.

8. In young gardens like those of the Myssovskaja Experiment Station with trees from 8 to 10 years old and of Professor Hrebizky, with trees 10 to 25 years old, a larger percentage of successful cross-pollinations may be observed than in old gardens. With the latter we should consider the garden at Djetskoje Sselo which is 40 to 50 years old.

9. In off years the percentages of success fall considerably. It is to be assumed that in such years the flower buds or the pollen or even both these elements are more impotent than in fruitful years.

10. Less cultivated and even wild forms seemed to be more utilizable for pollinations than highly cultivated varieties. The combinations of Pear with Tonkovjetka in the experiments of Belajev and others bear on such cases.

11. The capacity of the pear tree *Pyrus ussuriensis* to influence even the fruit development of apple varieties is notable.

12. As long as the most productive reciprocal pollinations are not completely established; it is most advisable to rely on mixed plantings of different varieties.

13. As an example of the most successful pairs of varieties for joint planting, as the experiments of the Pensa Horticultural School show, the apple varieties Longfield and the Transparent White should be considered. The latter is notable also on account of its high degree of self-fertility.

14. From now on the orchard should no longer be a chance conglomeration in which is heaped up varieties preferred for their external qualities or because of a gardener's caprice, but must present a harmoniously constructed, unfragmented unity, in which all individual trees are in close dependence upon one another and all collaborate together toward the attainment of their aim of highest productivity. The horticulturist, who plants a garden, should in this sense be an artist in his field.





# STERILITY IN THE STRAWBERRY AND ITS SOLUTION

GEORGE M. DARROW

*Bureau of Plant Industry, U. S. Department of Agriculture*

The problem of sterility in the strawberry has to do chiefly with impotence of the pistils of the hermaphrodite flowers of cultivated strawberries, and to a slight extent only, with abortive pollen. So far as we know, there is no self- or cross-incompatibility in this fruit as in the apple, pear, plum, cherry and other cultivated plants of the rose family. Though our problem may be less complex than that of the workers with these other plants it is of great economic importance. It is open to attack in two ways—genetically, by finding or breeding varieties which are completely fertile, or physiologically, by so modifying conditions for all strawberry growing regions that sterility would be suppressed. This paper reports some progress along each line.

In general, there are four groups of strawberry species from which cultivated varieties have been derived: *Fragaria vesca*, *F. elatior*, *F. chiloensis* and *F. virginiana*. Apparently, *F. vesca* and all of its allied species produce only hermaphrodite flowers and all flowers produced are fertile. There is no sterility known in this group of species. *F. elatior*, *F. chiloensis*, *F. virginiana* and allied species, however, are reported to have two flower types, pistillate which lack stamens, and hermaphrodite which are more or less sterile. Our own observations confirm this for the latter two species. *F. elatior* is a native of central Europe and we have not been able to observe this species to any extent. All important cultivated varieties, however, are derived entirely or chiefly from hybrids of *F. virginiana* and *F. chiloensis*. Varieties derived from *F. vesca* and *F. elatior* are small fruited and of little importance. Sterility in the strawberry is, therefore, of importance economically.

Genetically, the problem may be solved by crossing the species having no sterility and hermaphrodite flowers with our commercial varieties in order to transfer their complete fertility to cultivated sorts, to gradually breed out sterility, or to find varieties or wild selections which were entirely fertile.

Longley has found that *F. vesca* is a diploid, *F. elatior* is a hexaploid, and *F. chiloensis* and *F. virginiana* are octoploid chromosome species, the haploid number being seven. Intergroup hybrids in other crop plants have been found to be difficult to obtain and usually quite sterile. Richardson in England has reported similar results in crossing the species of strawberries. We have great difficulty in securing intergroup strawberry hybrids and those we secured were chiefly barren.

Longworth, over seventy-five years ago, reported that a low percentage of the flowers of hermaphrodite varieties set fruit while all flowers of pis-

tillate sorts set. In consequence, pistillate varieties were chiefly grown at that time. Since then a great increase in the fertility of cultivated sorts has occurred and on an average about 66 per cent of all flowers of all varieties set fruit at the Bell Horticultural Field Station in Maryland. It may, then, be possible to select the more fertile varieties for breeding and gradually raise the fertility. To this end, a record of the sterile flowers of many varieties is being secured for several years and the least sterile ones are being used in breeding.

In taking this record of the sterility of varieties one variety, the Rockhill, has been found which, so far, seems to be entirely fertile in all of its flowers. Crosses of this with other sorts indicates that this characteristic is inherited. Furthermore, the Rockhill variety possesses many desirable qualities which make it of value to breeders. It has very high quality, good size and fine appearance, the plants are very hardy and disease resistant. Apparently in this variety we may have the solution of the problem of sterility in the strawberry.

Schuster, in Oregon, has shown that the Ettersburg 121 strawberry grown for canning in Oregon and Washington is extremely productive on clay soils but quite sterile on sandy soils. Others have reported similar results. It is evident that, although the type of sterility in the strawberry is an hereditary character, in some varieties at least it is very sensitive to environmental influences. This leads us to hope that it can be modified in all commercial varieties to such an extent as to enable the grower to so manage his fields as to cause all flowers to set. To this end the type of sterility must be studied in detail and understood as far as possible.

Briefly, the fruit bud of the strawberry is laid down in the fall months in northeastern United States. As the fruit bud is terminal a vigorous strawberry plant to continue growth must do it through new lateral growing points which are found in the axils of the leaf blades. These new growing points develop into new crowns which in turn may form fruit buds at their terminals. The fruit buds develop rapidly and the pollen-mother-cell stage of flower development is apparently reached in a large part of the plants before winter. Ruef and Richey have shown that the primary flower of an inflorescence reaches this stage first, the two secondary ones next, the tertiary and later flowers successively later.

But sterility in an inflorescence appears in the same order as the development of flowers in all or nearly all varieties. The primary flowers are most often fertile, the secondary ones are less often fertile and the percentage of sterility increases in the later flowers. It might be suggested that the sterility of the pistils is due to a difference in the nutrition of the flowers, the primary flower blossoming first in the spring might take nutrients from the later flowers. However, we have picked the primary flowers of a selection which usually sets only its primary flowers and no change in the fertility of the later flowers resulted. Freezes often kill the pistils of primary flowers when they are very small buds. This has been observed on varieties the later

flowers of which are sterile and no change in fertility of such flowers was evident. Apparently, sterility of the pistils has already occurred before winter and is influenced or caused by fall conditions.

Records secured this spring on potted plants of the Dunlap variety grown in the greenhouse have shown greater sterility in the inflorescences on lateral crowns than in the inflorescence of the central crown. Similar results have been secured from records of multiple-crowned plants in the field. Apparently some condition more favorable to the production of fertile pistils occurs in the development of the earlier-formed fruit buds than in the later formed ones and it is to a study of conditions in the autumn that we should look for a physiological solution of the sterility of the strawberry.





# ABSCISSION IN GENERAL AND WITH SPECIAL REFERENCE TO THE CURTAILMENT OF FRUITAGE IN GOSSYPIUM

FRANCIS E. LLOYD  
*McGill University*

(WITH PLATE 13)

Among the causes which lead to sterility, using this term in a wide sense to mean non-fruitlet from whatever cause, and therefore curtailment of crop, are those which lead to the abscission of buds, flowers (style, petals, etc.), and fruit. These, however, are only special examples of a general phenomenon seen in the most diverse organisms, from Algæ to the Compositæ, which, with a few doubtful exceptions—as the cases of rhexolysis among the *Musci* (Correns, 1899) and of the style in *Gossypium* (Lloyd, 1914)—consists in the chemical alteration of the walls of the cells involved by oxidation (Sampson) hydrolysis, or other reaction. Something akin to the Canizzaryo reaction may take place, namely, the formation of an acid and an alcohol by the action of an alkali on an aldehyde. This possibility was indicated by my colleague, Professor H. Hibbert. The immediate activity of these cells (in metaphyta) is to secrete an agent (one or more) which acts on the outer regions of the wall and on the middle lamella, leaving unaltered a tertiary membrane, of various thickness in different organisms, clothing the still living cell. This, set free from its restraint, becomes rounded, and the turgor pressures set the self-imposed wound a-gaping (Wiesner, 1905). The release may be sudden and according to Fitting (1911), without cell-wall alteration the organ being vigorously expelled. The process is in essence the same in the algæ. I have recently studied it in *Mougeotia* and in *Spirogyra* amplifying somewhat the earlier studies of Strasburger (1875) and Benecke (1899). In *Spirogyra*, which undergoes abscission spontaneously when under untoward conditions (according to many observations), the first indication is the alteration of the external portion of the cell wall, the changes progressing inwardly till the inner zone of the wall in contact with the protoplasm is reached. This remains unaltered but can be stretched by the protoplasm. The wall around the end under the H-piece being similarly altered, the cell, in rounding up its now free end in response to turgor pressures, sets itself free. The middle lamella here is not altered so far as I have observed, but I imagine this to be due rather to the removal of the H-piece from the sphere of influence of the living protoplasm than to a program from which it is excluded.

*Mougeotia* presents a somewhat special case. In this plant the secondary membranes on both sides of the primary lamella become gelatinized in the course of events following cell division so that the transverse septum becomes lenticular. The primary lamella may tear; in many cases it is difficult to see it.

A dropping of turgor in a cell results in strains being set up which tear the cuticle opposite the septum, as observed by Benecke. The whole matter has been recently more fully described by myself (Proc. Mich. Acad. Sci. in press.).

The interest attaching to the above case of *Spirogyra* in the present connection lies in the practical identity of the process with that in the metaphyta, in which two programs of procedure are distinguishable, one in which abscission takes place with no preparation in the form of secondary growth of the abscission cells (*Syringa*, *Impatiens*, *Vitis*, *Ampelopsis*, etc.) and a second in which there takes place more or less growth as evidenced in cell division preceding, accompanying and even following the actual ptosis (*Coleus*, *Gossypium*, *Acer*, *Fraxinus*, *Mirabilis*, etc.). In all cases, separation results from alteration of the outer regions of the cell wall and of the middle lamella. The chemical action originates in the outer regions of the wall and extends progressively inwardly (toward the cell center) as shown by the relative avidity for specific stains (Ruthenium Red) of the different zones of material; but also by observing the advance of isotropicity of the altered wall. A very marked degree of swelling of the altered walls takes place in certain plants. Tison especially has emphasized this. In some cases, *Citrus*, for example (Hodgson, 1918), and as I have observed in *Euonymus* and *Populus*, the middle lamella and adjacent wall-staff resists alteration and remains for some time buried in the tumid walls, until it finally yields. Whether swelling of the walls is a marked feature of the process or not, the result is the same, the setting free of living separation cells.

If it takes place *around the whole cell at once*, there is a complete separation of that cell, which then may enlarge, become turgid independently and spherical—at once if the walls are thin enough, or later, as in *Polygonium Sieboldii*\* when the process of change in the wall has progressed sufficiently. If the process has engaged the entire abscission zone, ptosis of the organ now ensues. If, however, growth takes place and the abscission cells elongate and subdivide, the alteration of the wall proceeds at length within a zone around the cell roughly transversely with regard to the axis of the organ involved, permitting rapid longitudinal elongation of the cell. This is the "Schlauchzellmechanismus" of Löwi (1907) but it has always appeared to me that this term, while descriptive enough for some cases, notably *Mirabilis* as described by me, is too clumsy for adoption. The alteration of the wall may proceed around the end of the once mother-cell or between the newly formed daughter cells, or both, quite as in the other type, and thus the cells

---

\* Abscission of catkins takes place in this manner (Namikawa, 1926). Extensive zones of abscission in this mode occur also in the pericarp of *Aesculus*, between the operculum and perianth of the flower of *Eucalyptus*, between the valves of the fruit of *Oxalis*, etc. Fitting has inclined to view "chorism" in *Geranium* as a special case in which a sudden access of turgor causes the separation of the very small cells, arguing that the speed at which the response is obtained speaks against the action of enzymes. But it may not be assumed that no previous action of enzymes has taken place as indicated by the example of *Mougeotia*.

are set free and ptosis supervenes. Recently I. Namikawa has described abscission in *Malus communis*. The details of his account do not, I think, quite fit the facts as they have appeared to me from a study of *Malus* and of *Crataegus*, both of which fit the above second type perfectly. Of this, however, in another place. All living cells within the abscission zone behave similarly. Non-living elements, such as xylem vessels are set free at their faces of contact with living cells only; otherwise they remain unfreed. The more vessels there are, the more difficult is complete abscission. In the region of the vascular tissues, the separation layer tends to follow them upwards and in the presence of relatively old and more extensive wood, abscission pursues a course parallel to the vascular tissues and by dint of the application of growth turgor the wood is slowly and unevenly disarticulated. When morphological conditions of a peculiar kind, such as found in *Gossypium*, intervene, the process of abscission may take a most devious and extensive path. I refer to the fasciated condition often occurring between the pedicel and stem in the cotton (O. F. Cook, 1915; Lloyd, 1920).

Abscission, then, takes place only between living cells capable of a secondary growth response. It is in itself a quite normal occurrence, and, when it occurs to an unusual or abnormal extent, is a symptom of internal derangement. This is simply illustrated by the fact that a stem (if it can do so at all) will shed petioles from which the blades have been removed, but not if the blades are only injured (to what extent, however, we do not know). The obvious explanation here is malnutrition through the lack of photosynthates, and experiments and field observations (*Gossypium*, Harland) have thus been interpreted. But that this means merely lack of food materials does not seem to follow from the only attempt at quantitative study which has been made (Sampson, 1918). The abscission layer itself has, according to Sampson, the lowest amount of carbohydrates (reducing sugars) of any portion of the leaf or stem, and during the process of abscission the amount of nitrates increases. This latter observation would seem to harmonize with what we know, namely, that abscission is a vigorous growth process carried on by cells which have increased their protoplasmic machinery and are expending a large amount of energy. In experiments which I have done, in which malnutrition has a more than fair chance to come into play, I also have noted the presence in the vicinity of the mechanism of abscission of foods (starch) and their absence from the layer itself. Mason (1922) found that castrated flowers (*Gossypium*) do not shed so rapidly as usual if the other flowers and bolls of a branch are removed, and that the removal of the leaves of a plant is followed by total shedding. Though it would be easy to concede Mason's interpretation, that the limitation of the supply of assimilates is responsible by leading to negative growth rates, I venture to believe that in the case of boll-shedding they are not absent from the tissues immediately involved.

It is difficult to draw a clear conclusion with regard to the importance of food materials for abscission, by observing the amount of starch in or



near the separation layer since it may be nil or abundant. In the latter part of October both these extremes were illustrated by *Acer* and *Syringa* respectively. In the former, the tissues beneath the suberized layer were loaded heavily with starch, while only a trace was to be seen in the petiole above it, and this applies equally to the separation cells proper. When abscission overtakes the maple leaf, it is discolored and moribund, while the leaves of the lilac, in the separation cells of which there is an abundance of starch, fall while still green and not in the least discolored. However, the local enrichment of the separation cells in cytoplasmic (or similar) matter is quite obvious in both. Since in both separation had been achieved, the lack of immediately available food is not a necessary condition.

Whatever the amount of food required for abscission may be, the abscission layer itself cannot behave effectively in the absence of a plethora of water. A wilted plant does not procure abscission. I showed that field experiments with cotton begun in the evening were followed by shedding more rapidly than those begun in the morning, and this I attributed to a delay of response caused by the water deficit of an additional day. And Nami-kawa (1926) has afforded us some valuable measurements showing that the osmotic pressure of the separation cells "becomes remarkably high before the separation." Here Wiesner's views as to the rôle of turgor deserves recalling. If an early frost catches leaves before or during abscission, the process is stopped and marcescence prevails (as I observed in October, 1925) as it does also sometimes when a tree is badly affected by fungus lesions. In order to function, the abscission tissue must have, in addition to a source of energy, plenty of water; it is a growth response essentially.

Further evidence that abscission is a response of physiologically active cells is abundant although quantitative relations are difficult to determine. In a brief space it is not possible to review the results available from the studies of Fitting, Hannig, Goodspeed, McGee, Hodgson, Kendall (see literature cited by Kendall, 1918) and others. It is sufficient here to indicate the fact in its bearing on the above-mentioned thesis.

In many descriptions of abscission much attention has been paid to suberization or lignification, though to some authors the connection between the two processes is not regarded as inevitable or necessary (e. g., Heinicke, 1919). As a matter of fact it is not. In many plants lignification in the region of the separation layer (above it as well as below) occurs especially when the abscission falls at the close of the growing season. If procured at other times, and, at all events by a rapid response, no lignification takes place.

*Preformed abscission layer.* Although I have stated elsewhere that there is no morphological predetermination of the position of the abscission layer, a condition which is very common, it is, nevertheless, true that in some forms there are definite indications of differentiation of tissue to enable us to say definitely where abscission will take place. A striking example is the non-lignified single layer of cells beneath the flower of *Zizania*. Another is found in *Castilleja* which sheds massive woody primary branches as described by



F. A. F. C. Went (1897). Across the base of these branches a well-marked non-lignified layer may be seen, the only lignified elements traversing it being a few tracheids. In this layer, according to Went, the separation layer is formed in the manner described by Höhnelt as the usual sort of Von Mohl separation layer.

A predetermined abscission layer is not always marked by lignification but solely by the structural character of the zone of tissue itself, as, e. g., *Polygonum Sieboldii*. In the petiole base, but above the nectar gland, there is a constriction marking this zone, which consists of 3 or 4 layers of small isodiametric cells. It is here that abscission intervenes. Whether one will argue that this is, morphologically, an abscission layer will depend on the point of view. One interesting fact bearing on this is that in the climate of Montreal no abscission of leaves takes place till the close of the growing season. The leaves remain green, and no sign of abscission occurs till frost intervenes. Nevertheless, in twigs in a moist chamber abscission takes place in 4 or 5 days. I should rather argue that abscission occurs here where the cells have preserved their juvenude. That abscission happens in the same relative position in forms where there is no possibility of anatomical or morphological indications (e. g., *Ampelopsis*) is, on comparative grounds, an argument in support of the idea that the position of the separation layer is a physiological matter. It is because of their physiological peculiarities that the cells of the abscission region escape lignification in *Zizania* and *Castilleja*, one may argue, and these cells therefore are the only ones in which separation can occur. This is physiological predetermination, perhaps, and will be obnoxious to some, but nevertheless seems to be inescapable.

*Abscission in Gossypium.* Turning to consider specifically the case of the cotton plant we have for our information the work of Audebeau and of Balls and his associates in Egypt (ca. 1909 and on); of Orton and Duncan (in MSS. see Lloyd, 1920), and of Lloyd in Alabama, 1916, 1920, of Barre (1915) in North Carolina, of Ewing (1918) in Mississippi, and King in Arizona, all in the United States of America; of Nowell (Barbados, 1914), Harland (St. Vincent, 1919) and Mason (St. Kitts, 1922) in the West Indies; together with some work in India.

The mechanism of abscission is of the second type above mentioned. I have already described the matter elsewhere (1916, 1920). On this occasion I desire especially to indicate that there is no structural evidence of an abscission layer in the pedicel. Abscission may occur either above or below the groove which is evident near the base of the pedicel (Fig. 1). The course taken by the separation cells is usually as seen in Figure 1 dishing toward the supporting stem in the cortex and vascular region and upwards in the pith. But wide deviations from this rule and much irregularity may occur (Fig. 2), especially in decurrent pedicel bases.

During the preparation for separation there is a preliminary elongation of the cells involved, their subdivision once or twice, and chemical alteration of the longitudinal walls culminating in setting free the newly produced

cells from the old walls (Figs. 4-6). The whole fits precisely in all essential points the description of *Mirabilis* (Lloyd, 1916) and that type called by Löwi "Schlauchzellmechanismus."

Whether the pedicel is decurrent or not, the mode of abscission is the same, though it may under some circumstances lead to extreme histolysis (Lloyd, 1920).

The causes which have been regarded as responsible for the loss of buds ("squares"), flowers and fruits ("bolls") are: water stress in the plant whether due to soil or to high evaporation conditions, insufficiency of photosynthates, injury to the organ complex by fungi (Wolff, etc.) and by insects (boll weevil, pink bollworm, etc.) and inadequate pollination and fertilization, which, however, can apply only to young bolls which have refused to "set." Obviously the last named cause is restricted in time and experimentally determinable, and is so completely inapplicable to buds, open flowers and set bolls that we may dispose of it first. For the growing crop a sure, and perhaps the chief, cause of this condition is rain, which aside from its physiological effects as observed in St. Vincent by Mason, may be the cause of shedding by the destruction of pollen. This is recognized but does not play much part, naturally, where rain is scarce (Balls). I have shown that it can undoubtedly on occasions play a large part, but this also happens only when rain falls during the later morning hours since it would seem at this time that the stigmas are pollinated but that the pollen tubes have not yet carried the protoplasmic parts far into the style. Doubtless further study of temperature conditions especially would discover various time relations. Thus O. F. Cook states that when cool weather prevails the anthers do not open and fruit setting does not occur. The importance of ample pollination has been shown by Kearney (1921) and the very great complexity of the relation between development of the fruit (including seed) and pollination in *Zea* by Mangelsdorf (1926) who estimates that of twenty-seven genetic factors nine stimulate and eighteen retard various functions in the growing seed. It is therefore of importance in studying shedding where the rainfall is abundant that the time of the precipitation and its specific effect in the above indicated manner should be known. When it is not known the invaluable factor due to the abortion by rain can be suspected only, but the suspicion is enough to discount interpretation.

*Water stress in the plant leading to abscission.* It may be taken as settled that a widespread cause leading directly or indirectly to abscission of buds, flowers and bolls in the cotton plant is water stress. This plant, in common with others, experiences under most climatic conditions daily water deficits. Conditions in St. Vincent have been shown to work differently (Mason, 1922). Elsewhere, such water deficits may be amplified sufficiently to lead to abscission in the following ways.

1. Water shortage in the soil, exaggerated in the shallower layers by root interference. A gradual loss of water from the deeper soil reaches can produce an additive effect (Lloyd, 1920; King, 1922). The available water in

the surface soil may be modified by rains and by irrigation. If the rains are periodic, a corresponding periodicity of shedding has been observed (Lloyd); irrigations alternating with sufficiently extended periods of drought produce similar effects (Balls); and irrigation can stay an otherwise occurring wave of abscission (Barre).

2. The water stress initiated thus may be amplified also by above-soil conditions, such as excessive evaporation from the plant so that an epidemic of shedding may follow the incidence of a hot, dry wind (Balls). When cotton is widely spaced, the surface climate is less modified by the crop itself than when closely spaced, the latter condition preventing the effective influence of evaporation and so modifying shedding (Balls). With wide spacing, however, more small buds are shed than otherwise. Obviously intense isolation can be effective by promoting transpiration to too great an extent (Balls, Lloyd), though this is held not to occur in St. Vincent (Mason).

3. Too great a soil water content has been observed to effect abscission by asphyxiation of the roots as during the rise of the water table under irrigation (Balls), or resulting from the cooling effect of torrential rain in the tropics, in the opinion of Shepherd (see Mason, 1921, p. 196).

4. An otherwise sufficient soil water content can be deficient if the roots withdraw the water more rapidly than it can be replaced by capillarity, as suggested by Balls, and found to be probably the case in Arizona (King).

We may conclude without entering into further details that any circumstance or combination of circumstances in the environment which procures an undue water deficit in the cotton plant results in the shedding of buds and fruits to squares and bolls. "Open flowers" as well as "flowers" within three or four days after anthesis, i. e., "bolls only by courtesy"—are numerously shed in Egypt (Balls) but not elsewhere; indeed I have found indications of a definite inhibition of shedding during the period of anthesis (Lloyd). While water stress in the plant results in shedding, we are wholly vague if not ignorant as to how this works to stimulate abscission; for we know that abscission can take place in organs in the certain presence of a plethora of water (in the petioles of *Polygonium Sieboldii* under water, their blades removed) as Wiesner and myself have found. Two investigations (of Heinicke and of Mason) appear to throw some light on this. In St. Vincent the conditions of environment which make for shedding are apparently quite the reverse of those usually obtaining and are, namely, heavy rainfall accompanied by low evaporation, both conditions meaning also an overcast sky. Under such conditions, Mason believes that there ensues in the bolls an inadequacy of photosynthates which limits their water drawing power, leading to negative growth rates in the bolls and to abscission. Experiments involving removal of leaves support this view. Of abscission of petioles from which the blades have been removed and the reverse if unmolested (unless we assume the presence of some specific substance in the blade inhibitory of abscission) the only obvious explanation lies in a starvation effect, again difficult to admit in view of the repeated observation of starch,



etc., in the abscission layer (Goodspeed, Sampson, etc.). Mason's elucidation is based, however, rather on the insufficient amount of photosynthates in the mass of the boll and their consequent inability to draw in water. It is therefore the lack of water indicated by negative growth rates (again the general phenomenon of daily water deficit shown to obtain by Livingston and Brown, Lloyd, Shreve, Balls, etc.), which leads to abscission. Balls has argued that this, leading to closure of the stomata and consequent overheating of the boll, offers the stimulus.

Somewhat similar evidence is to be derived from the study by Heinicke of "June drop" in the apple, and the studies of Harvey and Murneek (1921) evidently have a similar import. Spurs on limbs with large leaves were found to lose their fruits less than those with small leaves; this was true also of strong spurs, i. e., those which have abundant storage tissue and are well provided with vascular tissues. More water passes to strong buds than to weak ones and the growth of the leaves proceeds accordingly. It was further found that seeds affect the osmotic value of the sap of the fruits, thus influencing the flow of sap. Heinecke, from both experiment and observation, concludes that "unfavorable conditions of food and water supply are among the basic factors which cause the dropping of flowers and young fruit." Without emphasizing minor deviations in the course of the argument it is clear that Mason and Heinicke take much the same view of the matter. Further the latter particularizes in saying that "abscission occurs when the movement of the sap into the pedicel through the separation layer" is inhibited by whatever means. Kendall has thought that it does not depend on the movement of food to the abscission zone, but I have found that the rate of abscission varies inversely with the length of the petiole (in *Polygonum Sieboldii*); though a piece of the petiole 1 mm. thick (including the abscission layer) will absciss, but takes longer than less curtailed petioles perhaps because of lack of nutrients.

We recall that Wiesner argued that summer leaf fall takes place in shaded leaves because of lowered photosynthesis following the smaller amount of light available after the astronomical midsummer. Brown and Escombe (1902) attributed leaf fall to the indirect effect of  $\text{CO}_2$  which disturbed the nutrient activities of the leaf.

No one therefore avoids the cessation of flow of sap, however caused, as the stimulus to abscission. None of the evidence seems to bring us much nearer to the heart of the problem, namely what it is which furnishes the immediate stimulus to abscission, a process which, as above pointed out, involves growth or at least energy expenditures by the abscission cells.

*Injury.* Injury may fall on the organ complex to be shed or the abscission layer itself. Probably the failure of certain gases or vapors to induce abscission can be traced to the too rapid and complete toxic effect on the abscission cells, just as early frosts procure marcescence by killing the cells of the abscission zone. This aside, therefore, injury of the shed part may be of a multitude of kinds and too many experiments have been done to be briefly



summarized (see Fitting, Hannig, Lloyd, Kendall, etc.), nor is it possible to draw very general conclusions, for many complications arise from circumstances of the extent, character of injury, and other conditions to which the parts are exposed. Fitting (1911), indeed, found that injury of the ovary inhibited the abscission of the petals in *Geranium pyrenaicum*.

Whether fertilization has occurred or not determines the effectiveness of injury; and injuries preventing fertilization are much more effective than those in similar or greater extent which do not, so that severe injuries by the boll-weevil to cotton flower-buds which do not castrate have no result, nor do injuries to bolls oftentimes, and the insect may even come to maturity within the ripened fruit. It is probably the extent and character of the lesion by this insect that determines the frequencies of shedding procured thereby, which are different from those of shedding due to abnormal water conditions (Lloyd, 1920). A totally unexplained phenomenon observed in the cotton square when attacked by the boll weevil, though not confined to this condition, is the flaring of the bracts. This appears to be hypernastic, at all events it can hardly be explained in relation to wilting though it is accompanied by chlorosis (Hunter and Pierce, 1912; Lloyd, 1920). Experimental attempts to induce this condition have failed me.

On the whole it seems reasonable to hold that lesions which affect the movement of sap in such a manner as to induce recurrent water deficit in the injured part are those which result in abscission. The lesions of fungi (which also are charged with responsibility for abscission, though exact observations are hard to come by) may work in the same way, though the excretion of toxic substances or other chemical agency may play a stimulatory rôle. But mere injury which does not affect the movements of sap are of very minor if not negligible importance (Wylie, Lloyd, Hannig).

The cotton plant presents an additional curious though not entirely exceptional behavior upon which some emphasis has been laid by O. F. Cook as a cause of abscission, namely, the disharmony of growth between the pedicel of the bud or fruit and the stem. The morphological conditions have been described (Cook, 1915; Lloyd, 1914, 1920). Briefly stated, there may occur more or less downward displacement of the pedicel base accompanied by longitudinal distortion of the same. Of the sequelae of this condition (at least in older bolls) is a continuity of the vascular tissues, due apparently to an early extension of non-living elements, so that bolls, which might normally be shed and actually fall away, suffer abscission indeed, but remain attached to the stem by strands of wood. The zone of abscission activity always follows the base of the pedicel, however distorted, and this sometimes leads to very extensive and irregular lesions, amply understood when it is realized that abscission activity is not confined to any particular layer or zone of "abscission cells" but tends to follow the vascular tissues longitudinally, the more the older these may be. Cook, in criticizing my work, believes, somewhat unjustly I think, that I have overlooked the morphological conditions (see Lloyd, 1916, 1920). He argues that the more rapid growth of the

stem as compared with the base of the pedicel produces a rupture of tissues, which he regards as abscission. "With sockets of the normal, circular or transversely elliptical form the pedicels often begin to tear loose at the base, while the buds and their enclosing involucre are still fresh and turgid. The buds may wilt after being partially detached or may fall with scarcely any wilting. It seems certain that the gradual reduction of the growth rates observed in cotton and in *Juglans* (Lloyd) correlated with water deficit, occurs before it can be observed except by experimental means. That the sockets are too large for the pedicel bases becomes apparent under a hand lens, since a gap is formed at once between the separating tissues of the pedicel and the socket, indicating a release of tension." (Cook, 1921, p. 200.)

This seems to say that abscission is a purely mechanical, passive response to tearing strains caused by the more rapid growth of the internode as compared with the transverse growth of the pedicel base. Now it must be insisted that in *Gossypium* abscission is always a growth process involving cell division. If we examine the abscission tissues we find furthermore that the course taken by the layer of active cells cannot be harmonized with passive tearing. In the third place, at the time when abscission is achieved there is no discrepancy of size as between pedicel-base and socket, and at all events there is no socket. The abscission layer may dish downwards but usually does not and there is, therefore, no depression in which the pedicel base fits at the time of abscission. The remaining portion of the pedicel forms rather a raised platform (Figs. 1-3). To examine an abscised pedicel with a hand lens after abscission is complete and the boll or bud loosened can tell nothing about relative sizes when abscission is effective.

I do not deny that strain due to disharmony of growth may not occur, but if so, such strain should be, and I believe is, greatest in cases of extreme decurrence of the pedicel. Nevertheless, many bolls hold and mature when the displacements are most pronounced. But the strain produced should tear the elongated pedicel base in a direction normal to that of the abscission layer. In fact this direction of tearing occurs (Lloyd, 1920) as shown by the occasional splitting of the stipule blade; and it can be observed readily on the earliest very short flowering shoots at the base of the cotton plant that the decurrent pedicel base has been ruptured normally to the axis of the stem, and not in the direction of the abscission layer; and if abscission occurs, it does so in the usual way. Further, sometimes an internode is caused to curve by the less actively growing pedicel base (Cook, 1915).

I have shown also that the shedding of buds in great numbers characterizes the brachytic variety *Dillon* (see also Cook, 1915), but these sheddings (not blasting *in situ*) occur without accompanying elongations of the internodes in a degree to produce differences of growth. The very fact that the variety is brachytic speaks against this. Again, on a branch of *Dillon* which I have before me I find on straight internodes—the mechanical conditions apparently most inimical to the successful adherence of the boll—four displaced and decurrent pedicels of which one only blasted, and such observations I

have repeatedly made. On the other hand much, if not the maximum, shedding occurs on very short internodes when still young and distorted by adhesion of the peduncle base (Cook, 1915) in which the mechanical stresses must be least effective.

I conclude therefore that, although there is shown by some varieties of cotton a greater susceptibility for shedding, this is not brought about by mere mechanical strains set up by disharmony of growth, but indirectly through disturbances in the distribution of sap and through greater competition for water or foods by juxtaposed numerous buds, and that abscission proceeds precisely as in normal varieties.

The competition of organs for water, especially when crowded on the axis is an observed fact (Mason, Hodgson, Lloyd), and it is probable that this plays a prominent role in the "cluster" cottons.

Nor must we overlook the possible bearings of facts brought to light by Harris and his associates (1924) with regard to the different salt content of Egyptian and Upland cotton—a higher chloride of the Egyptian type quite possibly meaning lower tendency to shedding. While these and other hereditary tendencies appear to play a role, as has been recognized by various workers (see Kearney and Peebles, 1926), I cannot agree with Cook that the curious morphological conditions seen in the cotton, especially in the "cluster" varieties can result in abscission in a purely mechanical way.

### SUMMARY

Abscission is a mode of activity of living cells whereby they separate from the tissue in which they occur.

1. The method is of two chief types: (a) Separation of cells *in situ* by chemical alteration of the middle lamella and adjacent secondary walls; and (b) Separation of cells produced by renewed meristematic activity followed by chemical changes in the walls of the same kind. At completion of separation the loosened cells are normally alive and capable of high turgidity. The quantitative studies of Manikawa are of interest here.

2. Abscission may or may not be accompanied by lignifications. There is no immediate or necessary connection between these processes, although, as a matter of fact, there is a sort of harmony of relation. This has not, however, been discussed.

3. The energy expended by abscission cells requires an immediately available food supply; and a plethora of water is also necessary. Lack of photosynthates or of water can work only indirectly, and by way of inducing conditions which are stimulatory to abscission.

4. Injury affects abscission similarly, except that both stimulatory and inhibitory influences have been detected.

5. That abscission in general is a response to some stimulus brings it into the same category as that to which the abscission of petals has been referred by Fitting. The particular details of the mechanism not yet being ascertained, it may be doubted that there is any distinction between the be-



havior of the petals studied by Fitting and other petals—I may cite, e. g., the corolla of *Gossypium*, which behaves as the first type above—or other organs.

6. Abscission in *Gossypium* is of the second type above mentioned. The various conditions known or supposed to lead to abscission of the buds, flowers and fruit in the field have been set forth, namely (a) the destruction of pollen by rain; (b) failure of anthers to open during low temperatures; (c) injury; (d) water stress, and (e) mechanical strains due to disharmony of growth between pedicel and stem.

7. Water stress may be procured both directly and by the lack of photosynthates due to insufficient light.

8. It is argued that the mechanical strains set up can have no direct effect on abscission, that this proceeds in the same fashion whether such strains are present or not.

#### LITERATURE CITED

- Balls, W. Lawrence, 1915. The cotton plant in Egypt. Macmillan & Co., Ltd., 1912.
- Balls, W. Lawrence, and Holton, F. S., 1915 *a*. The spacing experiment with Egyptian cotton. Phil. Trans. Roy. Soc. London, Series B. **206**: 107-80.
- 1915 *b*. The sowing-date experiment with Egyptian cotton, 1913. Phil. Trans. Roy. Soc. London, Series B. **206**: 403-480.
- Balls, W. Lawrence, and Holton, F. S., 1915 *a*. The spacing experiment with Egyptian cotton. Phil. Trans. Roy. Soc. London, Series B. **208**: 157-223.
- Barre, H. W., 1915-16. South Carolina Exp. Sta. Rep. **28** and **29**.
- Benecke, W., 1898. Mechanisms und Biologie des Zerfalles der Conjugatenfäden in die einzelnen Zellen. Jahrb. wiss. Bot. **32**: 453-476.
- Cook, O. F., 1915. Brachysm, a hereditary deformity of cotton and other plants. Jour. Agri. Res. **3**: 387-399.
- 1921. Causes of shedding in cotton. Jour. Heredity **12**: 199-204.
- Correns, C., 1899. Untersuchungen über die Vermehrung der Lambmose durch Brutorgane und Stecklinga. Jena.
- Fitting, H., 1911. Untersuchungen über die vorzeitige Entblätterung von Blüten. Jahrb. wiss. Bot. **49**: 187-263.
- Goodspeed, T. H., and Davidson, P., 1918. Controlled pollination in *Nicotiana*. U. Cal. Publ. Bot. **5**: 429-434.
- Goodspeed, T. H., McGee, J. M., and Hodgson, R. W., 1918. U. Cal. Publ. Bot. **5**: 439-450.
- Harland, S. C. Manurial experiments with Sea Island Cotton in St. Vincent, with some notes on factors affecting the yield. West Indian Bulletin No. 3.
- Harris, J. A., *et al.*, 1924. Jour. Agri. Res. **28**: 695-704.
- Harvey, E. M., and Murneek, A. E., 1921. The relation of carbohydrates and nitrogen to the behavior of apple spurs. Ore. Agri. Coll. Exp. Sta. Bull. 176.
- Heinicke, Arthur J., 1917. Factors influencing abscission of flowers and partially developed fruits of the apple (*Pyrus malus* L.). New York (Cornell) Agric. Exp. Sta. Bull. 393.
- Hodgson, R. W., 1918. An account of the mode of foliar abscission in citrus. Univ. Cal. Publ. Bot. **6**: 417-428.
- Hunter, W. D., and Pierce, W. D., 1912. Mexican cotton boll weevil. U. S. Bur. Entomol. Bull. 114.
- Kearney, T. H., 1921. Pollination of Pima cotton in relation to the yield of seed and fibre. Jour. Heredity **12**: 99-101.
- Kearney, T. H., and Peebles, R. H., 1926. Heritability of different rates of shedding in cotton. Jour. Agri. Res. **33**: 651-661.
- Kendall, John N., 1918. Abscission of flowers and fruits in the Solanaceae, with special reference to *Nicotiana*. Univ. Cal. Publ. Bot. **5**: 347-428.
- King, C. J., 1922. Water-stress behavior of Pima cotton in Arizona. U. S. Dept. Agr. Bull. 1018.
- Lloyd, F. E., 1914. Abscission. Ottawa Naturalist **38**: 41-52.
- 1916. Abscission in *Mirabilis Jalapa*. Bot. Gaz. **61**: 213-231.



- 1920. Environmental changes and their effect on boll-shedding in cotton (*Gossypium herbaceum*). *Annals N. Y. Acad. Sci.* **29**: 1-131.
- Löwi, E., 1907. Untersuchungen über die Blattablösung und verwandte Erscheinungen. *Ebenda*, **116**: 983-.
- Mason, T. G., 1920. Experimental work on cotton. Cotton Research Board, First Annual Report.
- 1921. A note on some recent researches on the cotton plant in the West Indies with special reference to St. Vincint. *West Indian Bulletin* **18**: No. 4, 184-197.
- 1922. Growth and abscission in Sea Island cotton. *Annals of Botany* **36**: 457-483.
- Namikawa, Isawo, 1922. Ueber die vorzeitige Abstossung der jungen Früchte von *Malus communis*. *Jour. Univ. Sapporo* **11**: (1) 1-21.
- 1922. Contributions to the knowledge of abscission and exfoliation of floral organs. *Jour. Col. Agri. Hoky. Imp. Univ.* **17**: (2) 63-131.
- Mangelsdorf, P. C., 1926. The genetics and morphology of some endosperm characters in maize. *Conn. Agri. Exp. Sta. Bull.* **279**.
- Sampson, H. C., 1918. Chemical changes accompanying abscission in *Coleus Blumei*. *Bot. Gaz.* **66**: 32-53.
- Strasburger, E., 1875. Ueber zellbildung and Zelltheilung. Jena.
- Went, F. A. F. C., 1897. Der Dimorphismus der Zweige von *Castilleja elastica*. *Ann. Jard. Bot. Buit.* **14**: 1-17.
- Wiesner, J., 1905. Ueber Frostlaubfall, etc. *Ber. Bot. Gesell.* **23**: 49-60.

## EXPLANATION OF PLATE 13

*Gossypium herbaceum*

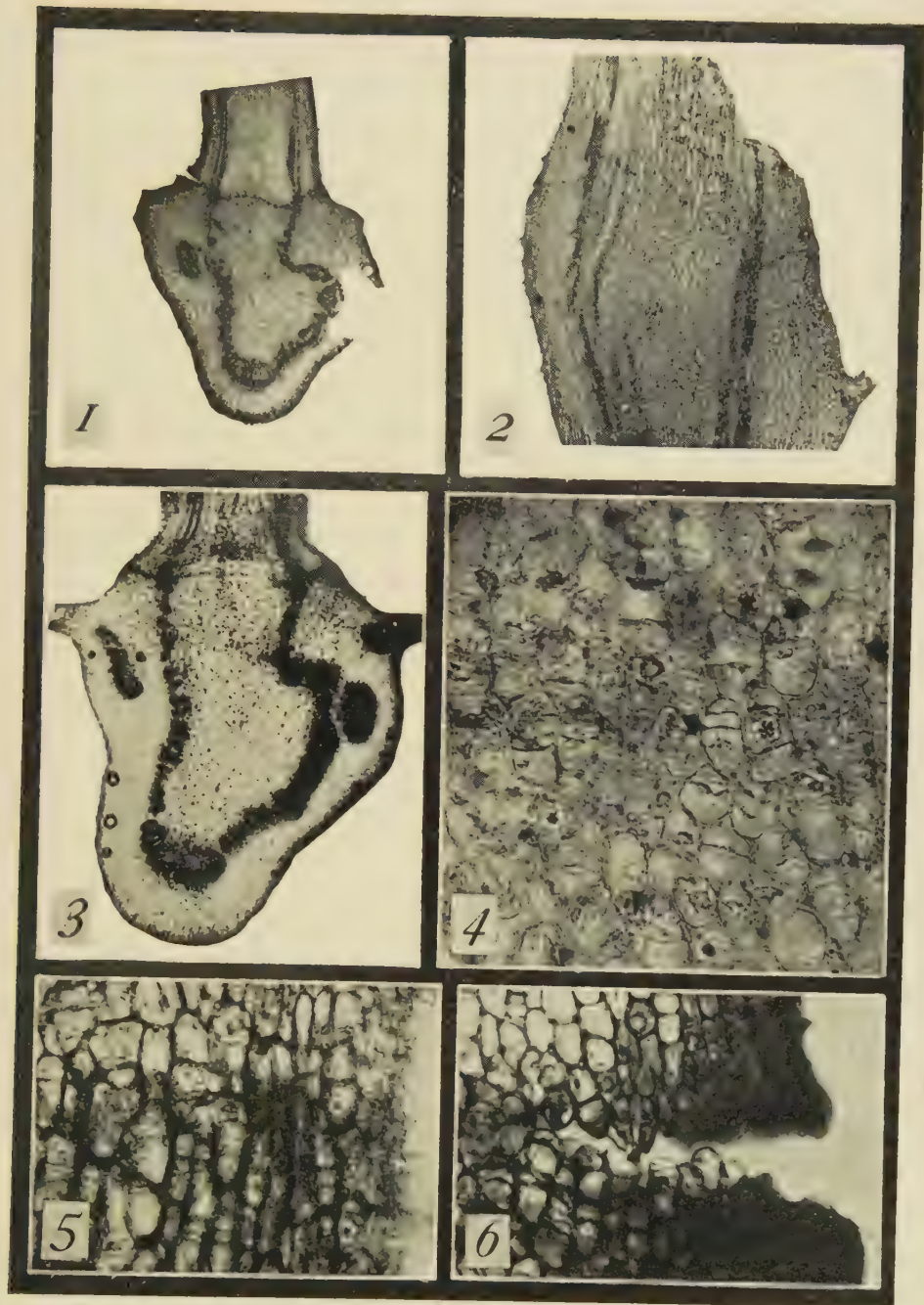
1-3 inclusive. Sections through junction of pedicel and stem of flower buds ("squares"). The pedicel is above in all the figures of the plate. The normal position of the abscission layer is seen in Fig. 1. In 2 a somewhat unusual case due to distortion of the parts is seen. In 3 the square had wilted and was shrunken and abscission was inhibited.

4. Abscission layer as it appears during the cell divisions preceding separation. Cortex.

5. The abscission layer in approximately the same position when the longitudinal walls are beginning to break down.

6. A similar case after separation has begun.





LLOYD: ABSCISSION IN GOSSYPIMUM





## STERILITY IN FILBERTS

C. E. SCHUSTER

*Oregon Agricultural College*

The results that will be outlined in this paper are from data secured during four years' work connected with the pollination needs of the commercial varieties of filberts as grown in the Pacific Northwest. Growers of filberts were at that time complaining that some plantings of filberts were either very light in bearing, or, as in some cases, were not bearing any nuts at all. A summary of the figures as obtained in that work are recorded in Bulletin 208 of the Oregon Experiment Station, and will not be given in this paper but only a general discussion of the same with related facts will be presented.

The filbert grown in the Northwest and Oregon is an introduction from Europe. Introductions were made from many different European countries, bringing with them a great deal of confusion as to varietal nomenclature. According to the best authorities the commercial varieties have been derived from the following European species: *Corylus avellana* L., *Corylus pontica* Koch, and *Corylus maxima* Mill., or as hybrids between two of these species. From observations of many seedlings raised from commercial varieties it would seem that these same commercial varieties have been derived as hybrids of the three European species commonly found, rather than that they are descended from one specie.

Occasional trees of *Corylus colurna* L. are to be found but these are of no economic value as yet. From the native hazel *Corylus californica* Rose, no cultivated variety has yet been developed. One named variety from the eastern form of wild hazel, *Corylus rostrata* Ait. has been tested but with negligible results.

In Oregon the main commercial variety is the Barcelona, a variety evidently introduced from Europe from several different countries and under several different names. The various introductions are now generally known as Barcelona, a name given this type of nut by Felix Gillette, the first extensive nurseryman and importer of filbert trees. The variety occupying a very secondary place is the Du Chilly. The Barcelona produces a round nut with a short husk that allows the nut to drop freely. The Du Chilly has a long nut that does not fall freely due to the long enveloping husk.

In the filbert, both types of flowers are borne on the same tree but usually do not mature at the same time. Under Northwest conditions the male flower or catkin is first noticeable about July 1, making a gradual growth from then on until just before blooming season when the greatest increase in size takes place. The female flowers of some varieties may be



## VEGETATIVE AND FRUITING BRANCHES IN THE DATE PALM AND STERILE INTERMEDIATES BETWEEN THEM

WALTER T. SWINGLE

*Bureau of Plant Industry, U. S. Department of Agriculture*

The Deglet Noor date variety, perhaps the best known date of North Africa and one of the finest dates that reaches the European and American markets, is now being grown on a large scale in the hot, irrigated valleys of Southeastern California and Southwestern Arizona.

This date variety like all other important varieties is propagated exclusively from offshoots which are vegetative branches that arise singly in the axils of the leaf when the palms are young. The young date palms show at the base a half dozen or more of such vegetative sprouts which are called offshoots by the date grower. When these offshoots have taken root they can be cut loose from the mother plant and transplanted without difficulty.

The very young date palm produces only vegetative buds, one in the axil of each leaf, but not all of these develop; many of them die when very young. As the palm gets older, say from three to eight years old, a part of the buds are vegetative and a part of them reproductive, that is, giving rise to flower clusters, female inflorescences of course in the case of a female variety like the Deglet Noor, since the date palm is a dioecious species. After the date palm reaches the age of eight or ten years, no more vegetative buds are produced in the axils of the leaves. All of the buds that develop give rise to inflorescences. So far as has been observed, after a date palm once begins to produce nothing but fruit buds, no more vegetative buds are formed. In other words, there is a slow change from producing entirely vegetative buds when young to a mixture of vegetative and reproductive buds after a few years and finally to a stage when only reproductive buds are formed, but this process is apparently irreversible, that is, no method of treatment yet discovered is able to force an old palm to produce vegetative buds.

After this short sketch of the normal development of the date palm which is shown by practically all varieties, both male and female, of *Phoenix dactylifera* and by some other species of *Phoenix*, it is interesting to note that in the case of the Deglet Noor date a very peculiar form of sterility exists.

This is due to what amounts in fact to a partial transformation of a vegetative bud to a fruit bud, or possibly the reverse, the partial transformation of a fruit bud into a vegetative bud. At any rate, the results of this imperfect expression of two different hereditary tendencies are very striking. A certain proportion, possibly 10 per cent, of the offshoots of the Deglet Noor palm are what the date growers call abortive offshoots. The leaves are abnormally slender and show few or no spines toward the base. The offshoot

is slender and weak and after a few years the terminal bud usually grows out into an abnormal female flower cluster and may often bear a few imperfect fruits, after which all further development of the abortive offshoot ceases and it finally dries up and dies.

This intermediate form of offshoot is much more like a vegetative offshoot than it is like a fruit cluster and is known to all careful observers of this variety of date palm. It is sometimes found in other varieties but the great majority of date varieties apparently do not show these abnormal abortive offshoots at all.

There is another even more striking form of intermediate development that, I believe, has not been recorded in the past, but which is very commonly found in the Deglet Noor palm; this is the abortive inflorescence. Certain buds give rise to a very peculiar, slender growth which is not enclosed in a spathe like the normal female inflorescence, but is more slender, more extended, and largely composed of abortive leaves. Such abortive inflorescences are frequently from eight inches to two feet or often three feet long and usually show very meager development of side branches and die within a few weeks after they appear. In the case of the Deglet Noor variety, these abortive offshoots often constitute a considerable proportion of the buds, particularly on inflorescences attached to young palms which have reached the fruiting stage.\*

To recapitulate then, we have here a new form of sterility, first, in the vegetative branches at the base of the Deglet Noor date palm leading to 10 per cent, or so, of these young offshoots developing abnormally and finally dying; in the second place, we have the formation on young date palms of a certain proportion, sometimes a large proportion, of buds which have some resemblance to inflorescences but which never produce normal flowers or fruit and which invariably die after a few weeks or a few months. In other words, by what amounts to a hybridization of the vegetative buds and fruiting buds we have the very curious phenomena of the production of sterile offshoots and sterile inflorescences through a mixture of the two conflicting hereditary properties in various proportions.

---

\* The thought has occurred to me that perhaps the fruiting palm is producing some hormone which diffuses down into the young offshoot at the base of the mother palm and so modifies their development as to cause the formation of such abnormal buds as have just been described.



## STERILITY IN PEACHES\*

C. H. CONNORS

*New Jersey Agricultural Experiment Station*

Horticultural varieties of peaches are usually propagated by budding so that seed production is not of vital importance, except as it concerns the production of new varieties. However, in the course of breeding operations at the New Jersey Agricultural Experiment Station, looking to the origination of new varieties, two types of sterility have been found: (1) *embryo abortion*, which is not of economic importance, and (2) *pollen sterility*, which is of great economic importance; with a possibility of a third type, pseudogamy.

### EMBRYO ABORTION IN PEACHES

In early attempts made at this station in the production of varieties of peaches that would mature early in the season, it was natural to use early varieties as parents. As little or nothing had been published on the behavior of peaches in breeding prior to this time, reciprocal crosses were made, using the early and late maturing varieties as parents. The method used was to cover entire trees with cheesecloth tents for the exclusion of insects and other factors of possible contamination.

Varieties which were used at various times were Mayflower (ripening at New Brunswick, N. J., in a normal season, about July 15), Early Wheeler or Red Bird (July 22), Arp Beauty (August 5), Greensboro (August 5), Carman (August 10), Miss Lola (August 12), St. John (August 12), Dewey (August 12), Slappey (August 15), Foster (August 20), Early Crawford (August 21), Fitzgerald (August 25), Belle (August 25), Elberta (September 3), Krummel (September 24), Heath (September 30).

Weather and other conditions permitting, fertilization takes place within 24 hours after pollination and the ovary begins at once to swell. Normally the fruit grows very rapidly up to the 44th day (by the 34th day the seed has reached its normal size but is filled with endosperm, the nucellus having almost disappeared) when the stone is outlined. By the 55th day the cotyledons have begun to develop and by the 61st day have about one-fifth filled the seed and the stones have reached maximum hardness.

The growth of the fruits of the variety Greensboro has been traced by measurements from pollination to maturity and the development of the seed has been macroscopically observed. This variety ripens in about 85 days from pollination. The gene, or whatever it is that governs the date of ripening, comes into influence, in the case of this variety, apparently somewhere between the 60th and 70th days, as from that time very rapid vegetative

---

\* Paper No. 313 of the Journal Series, New Jersey Agricultural Experiment Stations, Department of Horticulture.

development of the drupe takes place, and the fruit grows to maturity. When the fruit is mature, the embryo is well developed and the cotyledons likewise, although there is a difference in "texture," macroscopically, from the cotyledons of later maturing varieties.

In the case of varieties which ripen earlier than Greensboro (as Mayflower, Early Wheeler), the factor for earliness comes into influence sooner, as these varieties mature in 60 to 65 days and 70 to 75 days, respectively, from pollination. The cotyledons develop fully, apparently.

None of these varieties has produced any viable seed, either at Vineland, N. J., or at New Brunswick, N. J.

The next variety of commercial importance to ripen is Carman, which matures in 85 to 90 days from pollination. In this variety, the cotyledons develop in the fruits, and about 10 to 15 per cent of the fruits produce seeds which will germinate. This holds true of the other varieties ripening at about the same season, namely, Miss Lola, St. John and Dewey.

In varieties which follow this, there takes place a more definite retardation of growth of the drupe from about the 45th day up to 4 or 5 weeks before the date of maturity, during which period, enlargement of the drupe is very rapid. This longer retardation permits a fuller development of the embryo, and an increasing percentage of viable seed is formed. Slaphey will produce about 35 per cent viable seed. By the time the season of Belle is reached, and from then on, from 85 per cent to 95 per cent of the seeds are viable.

*Conclusion.* In the production of early varieties of peaches, it has been found impossible to use as seed parents varieties which ripen in less than 80 to 85 days from pollination, because the seeds will not germinate. When the fruit is mature, the cotyledons appear to be of normal size, usually, and in the case of Arp, an 85-day variety, the embryo appears to be well developed. The change, then, occurs after the fruit is mature. Since the early varieties are in a rapidly vegetative condition of the drupe at the time the drupes of the later maturing varieties are resting (vegetatively) and storing food in the cotyledons, accompanied by further development of the embryo, there appears to be a likelihood that the embryos of the early varieties are aborted before they reach the stage where the seeds will begin to germinate.

#### POLLEN-STERILITY

Varieties of peaches are, generally speaking, self-pollinating and self-fruitful, which indicates that the flowers are perfect and that no incompatibilities exist in fertilization. In the course of breeding operations and observation over a period of 14 years of nearly 200 commercial varieties and nearly 2,000 seedlings, however, varieties were found which failed to produce good crops under orchard conditions. The failure to produce crops in some instances was found to be due to pollen sterility.

In the breeding work each tree used as the seed parent was enclosed in a tent covered with cheesecloth. It has been our practice to emasculate and

pollinate by hand all the blossoms on about three-fourths of each tree so enclosed, and to allow the blossoms on the remaining one-fourth to be undisturbed. Among the varieties which have been tested are Mayflower, Early Wheeler, Greensboro, Arp, Carman, Miss Lola, St. John, Slappey, Foster, Early Crawford, Fitzgerald, Belle, Elberta, Krummel, Heath, Late Champion, Lippiatt's Red, Osprey Improved. Some of these varieties have produced better crops on the undisturbed portions than upon the hand pollinated parts. It must be noted, however, that some of the varieties used are very uncertain in bearing habits, even under orchard conditions. Foster, Fitzgerald and Early Crawford are particularly notable in this respect. These varieties belong to the so-called Persian group, many of the varieties of which are apt to start into growth in warm spells during the winter and hence are liable to winter injury of the blossoms, although several varieties of this group, no doubt, produce pollen sterile blossoms, as will be shown later.

The self-fertility of peach blossoms has been tested by many workers. Close (2) and Wiggans (9) bagged twigs of various important commercial varieties and found them to be self-fruitful. Coote (4) reported that in the greenhouse there appeared to be differences in the amount of pollen produced by various varieties of peaches in pots. He reports (and it is general knowledge among glass-house fruit growers) that peaches failed to set fruit unless pollinated with a brush or by bees. The failure to set fruit can probably be ascribed to lack of air currents in the greenhouse. While the wind is not believed to be a factor in carrying pollen, it probably plays an important part in moving or swaying the stamens so that the anthers come in contact with the stigmas. The same reason will probably hold true for the claim of Ewert (6) that self-fertility occurs sparingly among peaches. Ewert used potted plants of peaches for much of his work.

Under field conditions in this country only one case of sterility was reported prior to 1920. Fletcher (7) reported the variety Susquehanna as having a tendency to self-sterility.

### STERILITY OF SEEDLINGS

In the course of breeding operations with peaches in 1920, two seedlings of Belle self-pollinated were covered to secure the  $F_2$  generation. These two individuals had been several days later in blooming than most commercial varieties, a very desirable character, as a few days' delay in blooming may mean a crop. When these two individuals bloomed it was noticed that the anthers had a peculiar appearance in that they were small, very pale in color and when the period of dehiscence came no pollen was evident but the anthers turned to a brown color and shrank. Only a few fruits were set on each tree and as all blossoms were left to self the conclusion was reached that they were almost if not totally infertile as males. The occasional fruit was probably due to a few fertile anthers appearing in the flowers from which they developed.

During the next two seasons critical examinations were made of the



stamens of about 330 seedlings remaining in the fruiting blocks, about 500 undesirables having been removed, and of some 100 commercial varieties. It was found that J. H. Hale was the only commercial variety that had the peculiar condition of the anthers, although an occasional flower was found upon Slappey, Foster and Fitzgerald which were pollen sterile. Among the seedlings resulting from the breeding work, a number of individuals were found which were suspected of pollen sterility, as follows:

	Trees	Sterile	%
Belle, self-pollinated .....	157	33	21.0
Belle × Elberta .....	17	2	17.5
Elberta × Belle .....	27	4	14.8
Elberta, self-pollinated .....	46	6	13.5
Elberta × Early Crawford .....	14	1	7.1
Elberta × Greensboro .....	20	1	5.0
Belle × Greensboro .....	49	2	4.1

These percentages are based on the number of trees remaining after the undesirable had been removed. Probably all of the sterile sorts remain, so that on the total populations the percentages would be much smaller in some cases.

### STERILITY OF THE J. H. HALE PEACH

The J. H. Hale variety of peaches is so outstanding in its market and dessert qualities that as soon as it was offered for distribution it was widely planted in large blocks. Under orchard conditions, this variety of peach sets a great many much-undersized fruits, which have been called "buttons."

It was believed at first that these "buttons" were due to winter injury to the peduncle (Blake, et als., 1), or to improper fertilization (Palmer, 9). Parthenocarpic fruits have not been developed on the peach, after many trials by Ewert (5), but he did not work with this variety.

These "buttons" behave as if fertilization had occurred at least in part, for unpollinated blossoms drop very shortly after petal fall. The little fruits grow at a normal rate for about a month after blooming, and then growth is very slow. At the end of 80 days, the "buttons" measure about 19 mm. in diameter, while the normal fruits measure 32 to 38 mm. in diameter. At maturity, and the "buttons" mature a few days to a week later than the normal fruits, they are much undersized, poorly colored, flattened in shape and insipid in flavor. The stones are hard, nearly normal in shape but very small and with no kernel. The embryo appears to develop for 4 to 6 weeks and then ceases to grow.

What causes these "buttons" has not been determined, as none have appeared on the trees enclosed in tents for breeding or on other trees of the variety in the orchard under close observation. However, the J. H. Hale trees were few in number and were planted in such positions as to allow ample opportunity for cross-pollination. It may be due to winter injury to the peduncle, as suggested by Blake, et als. (1), who report buttons on Elberta, Foster, Fox and St. John. Most likely it is due to "improper fertilization" as suggested by Palmer (9), who reports "buttons" on Bilmeyer, Brigdon,



Early Crawford, Richmond and St. John. The pollen tube growth may be just great enough to cause an irritation to the ovule sufficient to start development. Probably fertilization does not take place and the development of the seed soon ceases. Possibly pollination does not take place until just after anthesis, when the ovules are just past condition for fertilization. The last is the most likely explanation.

The probability that J. H. Hale and the seedlings cited above were male sterile was deduced from observation. This was shown by germination tests. The pollen of self-fruitful varieties of peaches gives good germination on 5 per cent and 10 per cent sugar-agar media. Anthers of the J. H. Hale variety and of a number of the seedlings were dissected and crushed and placed in the media. Low-power microscopic examination showed that they were a mass of cells with no pollen grains present, hence no germination took place.

Following the announcement of the pollen sterility of the J. H. Hale variety of peach by Connors (3), Knowlton (8) undertook to determine the cause. He reports that irregularities in many of the tetraspores could be detected by January 1 and following liberation, abortion of the young microspores was frequent and continued until the period of dehiscence.

The tree habit and the fruit size, color, dessert quality and shipping quality of the J. H. Hale peach are outstanding and in spite of the pollen sterility of the blossoms and the susceptibility to collar injury in winter, it was deemed desirable to use it as a seed parent in breeding for better varieties of peaches. Accordingly, tents were erected over three trees in 1923, 1924 and over two trees in 1926. No emasculation was practiced after 1923, but each year at least one branch was left untouched while the blossoms on the remainder of the trees were pollinated by hand. Good sets of fruits were obtained when pollen of three commercial varieties and 21 of our own seedlings were used. In addition, pollen from two strains of *Amygdalus kansuensis*, and *Amygdalus communis*, as well as pollen from a double-flowered ornamental variety of *A. persica*, were used successfully.

On those branches the blossoms of which were not pollinated the following was the result: 1923, 756 blossoms, 0 fruit; 1924, 770 blossoms, 1 fruit; 1925, 593 blossoms, 5 fruits; 1926, 650 blossoms, 7 fruits; total, 2769 blossoms, 13 fruits. With most other varieties a good commercial set would have been obtained. The fact that occasional fruits are set, variable in number with season, indicates that under some environmental conditions J. H. Hale may be self-fruitful to a greater or lesser degree. Each year a cursory examination was made of all the blossoms left to self, and no anther was seen which produced pollen viable on media.

Under orchard conditions it has been recommended that J. H. Hale be interplanted with some other variety as a pollenizer. In one instance, in an established orchard, every fifth tree in every fifth row was top-worked to another variety. Following this, good uniform sets of fruits were obtained, with few "buttons." The peach is ordinarily not only self-fruitful but

autogamous, and in some seasons insects will play little part in pollination, due to weather conditions. There have been cases reported where solid blocks of J. H. Hale bore good crops without other varieties being adjacent, but inquiry showed that there was always an opportunity for cross-pollination. In one case the J. H. Hale was one-quarter mile on the windward side of a block of another variety. The bees were flying toward the Hale.

### MALE STERILITY IN CHINESE CLING

The variety Chinese Cling is the progenitor of many of our modern hardy commercial varieties, among them Greensboro, Carman, Belle and Elberta. Hiley is a seedling of Belle and J. H. Hale is doubtless a seedling of Elberta.

In 1924, a tree of Chinese Cling was covered and allowed to self, if it would, to see if Elberta (our most important commercial variety) and Belle would be reproduced. No fruits set. In 1926, the tree was covered again and one branch was left untouched. An examination of the blossoms showed the same condition of anthers and pollen as obtained in the J. H. Hale variety and in the seedlings mentioned above. On this appeared about 250 blossoms, from which 4 fruits developed. The remainder of the tree was hand pollinated with pollen from two commercial varieties, Early Crawford and Oldmixon and a good set of fruits was obtained. This is conclusive evidence of the pollen sterility of this sort, and gives a clue to the possible origin of the pollen sterility of seedlings of Belle and Elberta, including J. H. Hale.

About 2,000 seedlings of J. H. Hale from controlled crosses are now available for study, and will fruit within the next two or three years. The only material available for immediate study are 127 seedlings of J. H. Hale from open fertilized seeds. The parentage of these is not known but it will no doubt be possible to tell, after fruiting, as the varieties possible for pollination are known.

Of these 127 seedlings, 42 or 33 per cent appeared to be sterile, judging by the condition of the anthers.

### SUMMARY

In the peach are found two types of sterility:

1. Embryo abortion is present in those varieties which ripen within about 90 to 100 days from the time of blooming. Those varieties which ripen in 75 to 80 days produce no viable seeds, those which ripen in 85 to 90 days produce only 15 per cent viable seeds and those which require a longer period for the maturity of the fruits produce from 50 to 85 per cent of viable seeds. The stones are well formed, though sometimes insufficient quantities of stone cells are laid down to give complete hardness. Except in the very earliest varieties the cotyledons are well formed.

The probable cause of this abortion is the fact that enlargement of the drupe takes place at the same time as the final period of development of the embryo, due to a heritable character determining the period of ripening. The

flesh becomes mature and ripe and abscission takes place before the development of the embryo is completed.

2. Pollen sterility has been exhibited in seedlings resulting from our breeding work and has been found in several commercial varieties, notably Chinese Cling, the stem parent of many of our commercial sorts, and J. H. Hale and in addition occasionally in Late Crawford, Elberta, Belle, St. John, Early Crawford.

Pollen sterility is evidenced in the appearance of the stamens. The filaments are of usual length but the anthers are pale in color and fail to dehisce, instead drying up. The anthers are filled with a shrunken mass of broken down tissue. According to Knowlton, the abortion takes place after the tetraspore stage.

The remedy for this condition, in the orchard, is interplanting with other varieties as pollenizers.

Of 127 seedlings from open pollinated fruits of J. H. Hale, 42 or 33 per cent are pollen sterile, judging by the appearance of the stamens.

When trees are covered by tents for breeding work, branches are always left to self-pollinate. Most commercial varieties set good crops of fruit without artificial pollination. In tenting experiments the J. H. Hale over a period of 4 years has set 13 fruits out of a total of about 2800 blossoms.

Chinese Cling in 1926, set 4 fruits out of about 250 blossoms on a tree under cover.

3. The formation of "buttons" on the J. H. Hale and other varieties may be a third type, pseudo-gamy.

#### BIBLIOGRAPHY

1. Blake, M. A., Farley, A. J., and Connors, C. H. Report of the Horticulturist. N. J. Agric. Exp. Sta. 38th Ann. Rept. 1917: 119.
2. Close, C. P. Report of the Horticulturist. Del. Agric. Exp. Sta. Rept. 1903: 117.
3. Connors, C. H. Fruit setting on the J. H. Hale peach. N. J. Agri. Exp. Sta. 43rd Ann. Rept. 1922: 102.
4. Coote, George. Notes on the comparative date of blooming and pollen production of varieties of apples, pears, plums and cherries. Ore. Agric. Sta. Bull. 34. 1895.
5. Ewert, R. Recent investigations on the occurrence of parthenogenesis among orchard trees and other fruitbearing plants. (Trans. title.) Landw. Jahrb. 38: 767-839. 1909.
6. Ewert, R. A new case of parthenocarpus development. (Trans. title.) Jahresber. Agew. Bot. 5: 83-85. 1907.
7. Fletcher, S. W. Pollination in the orchard. N. Y. (Cornell) Agric. Exp. Sta. Bull. 181. 1900.
8. Knowlton, W. E. Pollen abortion in the peach. Proc. Amer. Soc. Hort. Sci. 1924: 67-69. 1925.
9. Palmer, E. F. Report of the Horticultural Experiment Sta. Ontario Dept. Agric. 1919: 43.
10. Wiggans, C. C. Progress report on horticultural investigations. (Report of the director of the Mo. Agric. Exp. Sta. for 1914.) Mo. Agric. Exp. Sta. Bull. 131. 1915.

autogamous, and in some seasons insects will play little part in pollination, due to weather conditions. There have been cases reported where solid blocks of J. H. Hale bore good crops without other varieties being adjacent, but inquiry showed that there was always an opportunity for cross-pollination. In one case the J. H. Hale was one-quarter mile on the windward side of a block of another variety. The bees were flying toward the Hale.

### MALE STERILITY IN CHINESE CLING

The variety Chinese Cling is the progenitor of many of our modern hardy commercial varieties, among them Greensboro, Carman, Belle and Elberta. Hiley is a seedling of Belle and J. H. Hale is doubtless a seedling of Elberta.

In 1924, a tree of Chinese Cling was covered and allowed to self, if it would, to see if Elberta (our most important commercial variety) and Belle would be reproduced. No fruits set. In 1926, the tree was covered again and one branch was left untouched. An examination of the blossoms showed the same condition of anthers and pollen as obtained in the J. H. Hale variety and in the seedlings mentioned above. On this appeared about 250 blossoms, from which 4 fruits developed. The remainder of the tree was hand pollinated with pollen from two commercial varieties, Early Crawford and Oldmixon and a good set of fruits was obtained. This is conclusive evidence of the pollen sterility of this sort, and gives a clue to the possible origin of the pollen sterility of seedlings of Belle and Elberta, including J. H. Hale.

About 2,000 seedlings of J. H. Hale from controlled crosses are now available for study, and will fruit within the next two or three years. The only material available for immediate study are 127 seedlings of J. H. Hale from open fertilized seeds. The parentage of these is not known but it will no doubt be possible to tell, after fruiting, as the varieties possible for pollination are known.

Of these 127 seedlings, 42 or 33 per cent appeared to be sterile, judging by the condition of the anthers.

### SUMMARY

In the peach are found two types of sterility:

1. Embryo abortion is present in those varieties which ripen within about 90 to 100 days from the time of blooming. Those varieties which ripen in 75 to 80 days produce no viable seeds, those which ripen in 85 to 90 days produce only 15 per cent viable seeds and those which require a longer period for the maturity of the fruits produce from 50 to 85 per cent of viable seeds. The stones are well formed, though sometimes insufficient quantities of stone cells are laid down to give complete hardness. Except in the very earliest varieties the cotyledons are well formed.

The probable cause of this abortion is the fact that enlargement of the drupe takes place at the same time as the final period of development of the embryo, due to a heritable character determining the period of ripening. The



flesh becomes mature and ripe and abscission takes place before the development of the embryo is completed.

2. Pollen sterility has been exhibited in seedlings resulting from our breeding work and has been found in several commercial varieties, notably Chinese Cling, the stem parent of many of our commercial sorts, and J. H. Hale and in addition occasionally in Late Crawford, Elberta, Belle, St. John, Early Crawford.

Pollen sterility is evidenced in the appearance of the stamens. The filaments are of usual length but the anthers are pale in color and fail to dehisce, instead drying up. The anthers are filled with a shrunken mass of broken down tissue. According to Knowlton, the abortion takes place after the tetraspore stage.

The remedy for this condition, in the orchard, is interplanting with other varieties as pollenizers.

Of 127 seedlings from open pollinated fruits of J. H. Hale, 42 or 33 per cent are pollen sterile, judging by the appearance of the stamens.

When trees are covered by tents for breeding work, branches are always left to self-pollinate. Most commercial varieties set good crops of fruit without artificial pollination. In tenting experiments the J. H. Hale over a period of 4 years has set 13 fruits out of a total of about 2800 blossoms.

Chinese Cling in 1926, set 4 fruits out of about 250 blossoms on a tree under cover.

3. The formation of "buttons" on the J. H. Hale and other varieties may be a third type, pseudo-gamy.

#### BIBLIOGRAPHY

1. Blake, M. A., Farley, A. J., and Connors, C. H. Report of the Horticulturist. N. J. Agric. Exp. Sta. 38th Ann. Rept. **1917**: 119.
2. Close, C. P. Report of the Horticulturist. Del. Agric. Exp. Sta. Rept. **1903**: 117.
3. Connors, C. H. Fruit setting on the J. H. Hale peach. N. J. Agri. Exp. Sta. 43rd Ann. Rept. **1922**: 102.
4. Coote, George. Notes on the comparative date of blooming and pollen production of varieties of apples, pears, plums and cherries. Ore. Agric. Sta. Bull. 34. 1895.
5. Ewert, R. Recent investigations on the occurrence of parthenogenesis among orchard trees and other fruitbearing plants. (Trans. title.) Landw. Jahrb. **38**: 767-839. 1909.
6. Ewert, R. A new case of parthenocarpus development. (Trans. title.) Jahresber. Agew. Bot. **5**: 83-85. 1907.
7. Fletcher, S. W. Pollination in the orchard. N. Y. (Cornell) Agric. Exp. Sta. Bull. 181. 1900.
8. Knowlton, W. E. Pollen abortion in the peach. Proc. Amer. Soc. Hort. Sci. **1924**: 67-69. 1925.
9. Palmer, E. F. Report of the Horticultural Experiment Sta. Ontario Dept. Agric. **1919**: 43.
10. Wiggans, C. C. Progress report on horticultural investigations. (Report of the director of the Mo. Agric. Exp. Sta. for 1914.) Mo. Agric. Exp. Sta. Bull. 131. 1915.



# STERILITY IN CERTAIN GRAPES

TORASABURO SUSA

*Hokkaido Imperial University*

(WITH PLATE 14)

## STERILITY IN MADELAINE ANGEVINE IN NORTHERN JAPAN

This variety belongs to the species of *Vitis vinifera* and has the imperfect hermaphrodite type of flower. Although its fruit ripens early in season it flowers late in spring. This variety was imported from France to Japan about forty years ago. For the first twenty years, however, it was not cultivated but on account of its excellent characters, hardiness in growth, good quality of fruit and especially in its productivity and early ripening habit, it has become one of the important commercial varieties of northern Japan.

But very generally throughout the areas of its cultivation complaints came from the growers regarding the "coulure" or the poor setting of fruit by this variety. This led to the studies which the author made in 1922 and 1923 on the character of the flowers of this variety and the requirements for the setting of fruit.

The investigation was mainly performed in the vineyard, greenhouse and in the laboratory at the Horticultural Department, Agricultural College, Hokkaido Imperial University, Japan. For comparison studies were made at the Niizuma vineyard, a commercial vineyard which is located along the Toyohira River just outside the city of Sapporo.

There are three main flower types in the cultivated seeded grapes of Japan, including both *vinifera* varieties and varieties from Northeastern United States. These types are as follows:

A. Perfect hermaphrodite flower: a bottle-shaped normal pistil and long upright stamens, both of which are highly functional (1 in FIG. 1).

B. Imperfect or pseudo-hermaphrodite flower: a normal pistil and semi-reflexed stamens (2 in FIG. 1).

C. Imperfect or pseudo-hermaphrodite flower: a normal pistil and very short and reflexed stamens (3 in FIG. 1).

According to Hedrick\* out of 132 of the important commercial grapes 97 have upright stamens and 37 have reflexed ones. As noted by Stout† there are in grapes numerous gradations between the flowers with fully upright stamens and the flowers with extremely reflexed stamens. The type B noted above is one of these intermediate types.

The author observed in Japan (Iwanohara) that eight cultivated varieties had the B type (semi-reflexed stamens) and nine had C type of flower out

---

\* Manual of American Grape Growing. 1919.

† Types of flowers and intersexes in grapes. Technical Bulletin No. 82. N. Y. Agric. Exp. Sta. 1921.

of seventeen pseudo-hermaphroditic varieties. None of these varieties yielded pollen that was viable in germination tests on the artificial media which gave excellent germination of the pollen of varieties having the perfect hermaphrodite type of flower.

The flower type of Madelaine Angevine belongs to *B* sort, above mentioned, and its pollen has shown no viability in several germination tests on the artificial media, which was carefully made up with redistilled water and 10 and 20 per cent sucrose, and which previously showed best results for normal grape pollens. The pollen of Madelaine Angevine is apparently impotent. It is very irregular in size and shape with many small and misshaped grains compared with other perfect hermaphrodite varieties. This

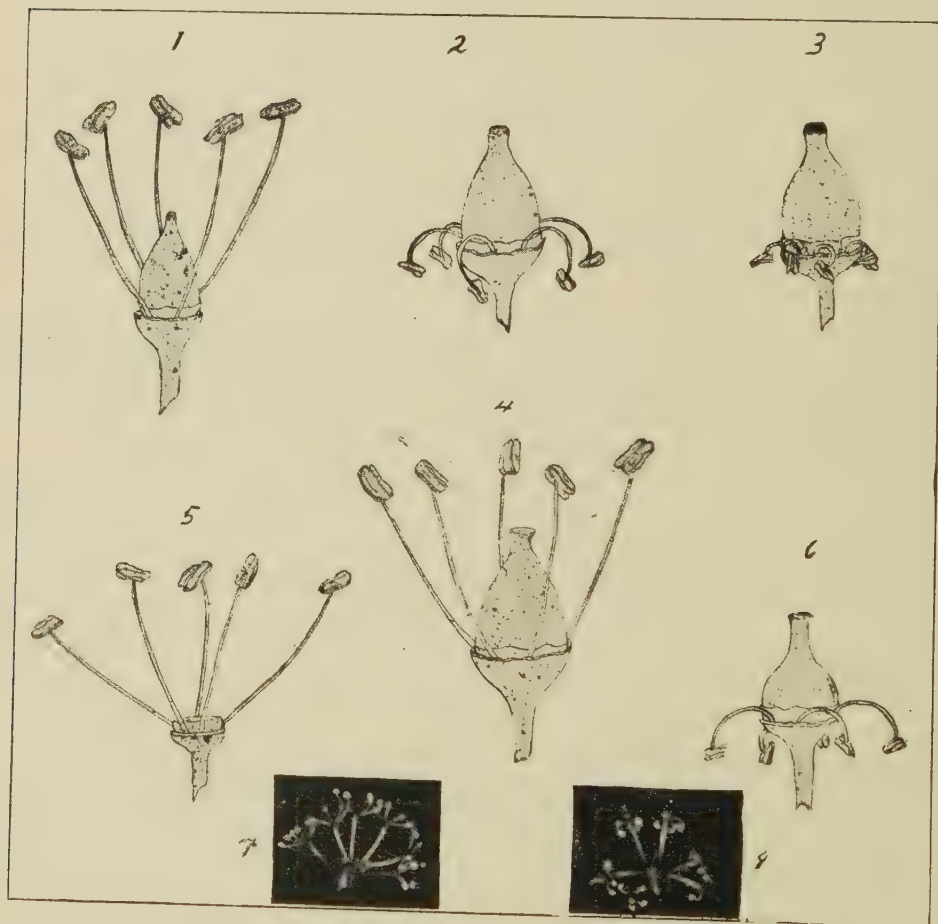


FIG. 1—Types of flowers in grapes:

1. Perfect hermaphrodite flower of the Delaware variety.
2. Imperfect hermaphrodite of type B from Madelaine Angevine.
3. Imperfect hermaphrodite of type C from Lindley.
4. Flower from the seedless White Corinth. Appears similar to perfect hermaphrodite but is not able to produce seeds.
- 5 and 7. Staminate flowers from a male vine of the wild Japanese grape *Vitis Coignetiae* and
- 6 and 8. Imperfect flowers from another plant.



variety, however, in its self-pollination tests, especially of plants growing in the greenhouse, has shown some self-fertilization.

Ten flower clusters in the greenhouse and twenty in the vineyard were bagged before blooming for self-pollination. When the flowering time came the bags were shaken by hand several times and the bags left until the ripening time. In the greenhouse tests, one to eight semi-normal seeds developed in each cluster which formed with variously sized berries numbering twenty to one hundred and nine to the cluster (See 1, PLATE 14). The size of the berries was apparently correlated with the seed development. In the outside tests, however, there were very few berries formed to self-pollination. In the university vineyard, which is rich in humus, only a very few small parthenocarpic fruits developed in the sixteen clusters self-pollinated and these contained no fully developed seeds. Similar results were obtained in a commercial vineyard.

Therefore some pollen of this variety may have influence for the fertilization in spite of its general imperfect appearance which might have varied with environmental conditions for pollen development as well as for pistil function. But this variety is practically self-fruitless due to the poor pollen which it produces.

The remedy for the self-fruitlessness of such a variety as Madelaine Angevine is sufficient interplanting with a variety which yields potent pollen and has the same blooming period. These pollenizers should also yield fruit that is of commercial value.

The flowering time of Madelaine Angevine at Sapporo comes quite late, about July 20, and the stigmas change into black color after one week at which time fruits of most of the American varieties have already grown to about the size of peas. In spite of this late-blooming habit, the fruit of Madelaine Angevine ripens early, about August 25 to 30, while the fruits of Delaware are not ripe until about September 15 to 20. The variety Beacon which blooms about the same time as Madelaine Angevine ripens fruit about October 1st.

Varieties blooming at Sapporo at the same time as the Madelaine Angevine variety are Pinot Blanc, Precoce de Malingre, Beacon. Blooming somewhat earlier are Pinot Gris and Delaware. As the best pollenizer for Madelaine Angevine, the first recommendation is given to the Precoce de Malingre and the second to Pinot Blanc according to their commercial value for such temperate zone.

Sterility of the variety Madelaine Angevine, therefore, is due chiefly to its pollen impotence. To secure full fruitfulness one should interplant with some of above mentioned varieties and it is recommended that this be done for every third row of the vineyard.

#### STERILITY IN SEEDLESS GRAPES WITH REFERENCE TO FRUIT FORMATION

In this study the author has tried to determine what the conditions are that lead to the development of fruits on seedless grapes, i.e., whether there

is development of fruits without pollination and fertilization (autonomic parthenocarpy) or whether the stimulus of pollination and fertilization is necessary (aitonomic parthenocarpy).

This investigation was conducted (1) at the Iwanohara commercial vineyard located about two hundred miles northwest from Tokio and about one thousand miles southwest from Sapporo, (2) at Tanaka vineyard which is located about one hundred and fifty miles westward from Sapporo in the Hokkaido Island and (3) at the Horticultural Laboratory, Hokkaido Imperial University, all during the year of 1922 and 1923 and then (4) later in 1924 at the Viticultural Department, University Farm, Davis, California.

For the first year the author went to the Iwanohara vineyard and tried to study the three seedless varieties, White Corinth, Sultanina and Seedless Sultana by bagging with paraffin paper bags before flowers open and leaving the bags intact until the berries ripen. At the same time the pollens of those grapes were collected into vials and sent to the Sapporo Laboratory, being kept in the dessicator using calcium chloride during the two weeks journey. As soon as the pollens reached the laboratory, germination tests were made by the hanging drop method of sugar solution and by sugar agar media.

Then the second year the stamens were removed a few days before the flowers opened. Careful attention was taken to remove all stamens left in the cluster. The emasculated clusters of flowers were enclosed in translucent paraffin paper bags and left for the ripening season.

It must be remembered that to castrate these grape flowers is a very difficult procedure. The earlier the season the more harmful for the flower. If it were too early, e.g., too young in bud structure, the cutting of the caps and stamens kills the entire flower cluster, especially if done in dry weather.

In Japan only three seedless grapes are grown. These are the White Corinth, Thompson seedless (Sultanina) and Seedless Sultana. At the present time such other varieties as Black Monukka, Sultanina rosea, Sultanina giant and Black Corinth are not grown in Japan.

In general appearance the flowers of all these seedless grapes closely resemble the perfect hermaphrodite types (4, in FIG. 1). They have a pistil which appears to be of normal form. The stamens are upright and an abundance of viable pollen is produced. In the seedless Sultana the pistil is somewhat smaller than in the other varieties.

Germination tests were made in Japan for pollen of White Corinth, Sultana and Thompson Seedless. The medium used was a 20 per cent sugar solution in redistilled water at a temperature of 25° C., and a duration of 24 hours. For White Corinth from 10 to 20 per cent of the grains germinated and for the Thompson Seedless 20 per cent germinated, all giving long tubes. No germination was obtained with pollen of Sultana but only one lot of pollen was tested and this was not tested until 16 days after collection.

In America at the vineyards of the University of California, germination tests were made of freshly collected pollen of Black Monukka, Black Corinth, White Corinth, Sultana, Thompson Seedless, Sultanina rosea and Sultanina

giant. The lowest percentage of germination was 30 for pollen of White Corinth; the highest was 80 per cent for pollen of Sultanina rosea. In every case the pollen was rather highly viable and the pollen tubes were long.

In 1922 carefully controlled self-pollinations of the seedless grapes White Corinth, Thompson Seedless and Sultana showed that they were all fully self-fruitful. Cross-pollinations with the highly viable pollen of the varieties Carignone and Folle Blanche gave no noticeable increase in fruit production over self-pollinations.

In the following year tests were made to determine if these seedless grapes will set fruit without any pollination, that is, may their fruits be purely parthenocarpic? The flowers of seven clusters on White Corinth were emasculated and enclosed in transparent paper bags. A few days later at the time when this variety was shedding pollen, three of the clusters were cross-pollinated with pollen of another variety known to produce highly viable pollen and the rest of four were not pollinated at all. All seven clusters remained enclosed in the paper bags until September. The four emasculated and unpollinated clusters set fruit as well as did the three cross-pollinated. In no case were fully developed seeds present. This seems to indicate that the White Corinth may produce fruit that is parthenocarpic. In 1924 similar tests were made at the experiment stations at Davis, California with Black Monukka and Sultanina. Five flower clusters of Sultanina were emasculated in buds and enclosed in paper bags. Of these three were destroyed by wind. In the other two, of 222 flowers 30 berries were developing and of 386 flowers 97 berries were maturing on August 7. The berries were all below the normal size for this variety. Three clusters of flowers were bagged without emasculation and thus subjected to self-pollination. One cluster was destroyed by wind. The other two set 205 berries out of 310 flowers and 395 berries out of 840 flowers. These berries were of the sizes normal for the variety. Quite the same results were obtained for Black Monukka.

According to the author's observation, there are various grades of seedlessness in so-called seedless grapes. The author had observed what appeared to be well developed seeds in fruits of the seedless Sultana and White Corinth. In the former variety the berries may be produced when selfed which contain such rudimentary seeds as well as open crossed, but in the White Corinth there is entirely no such seed development as the writer had observed. This imperfect seed development was just like when selfed in Madelaine Angevine.

As the pollens of those seedless grapes is highly viable, the various grades of seedlessness may be due to the various grades of abortiveness in embryo. On the contrary, in the Madelaine Angevine, its seedlessness from self-pollination is evidently due to imperfect pollination.

In many of the seeded grapes undersized berries with aborted or rudimentary seeds are somewhat frequent apparently due to imperfect fertilization.



The seedless grape varieties which were studied all produce pollen that is highly viable in artificial germination. The evidence indicates that White Corinth will produce fruit without pollination or fertilization. Sultanina will also produce such fruits but evidently these are smaller than the fruits which contain rudimentary seeds.

#### STERILITY IN THE JAPANESE WILD GRAPE, *VITIS COIGNETIAE* PULLIAT

This species is indigenous in Japan, especially thriving in the temperate zone. In the Hokkaido Island it can be seen almost everywhere over the plains as well as in the woods and mountains. It has a very strong-growing vine, covering trees and arbors with a thatch of heavy showy foliage, branches floccose-tomentose when young; tendrils intermittent; leaves cordate-orbicular with 3 to 5 lobe-like points, the margins apiculate-toothed shallowly dull above, thickly gray-pubescent beneath; thyrses stalked short; fruit late-ripening globular, about 1.2 cm. diam., purple-black color with white-bloom when ripe; pulp juicy, detachable from the skin, reddish-purple, sweet, good quality; flower very early, 2 types, staminate and pistillate by the vine. (See 5, 6, 7, 8, FIG. 1.)

This species has two types of flowers. One is the staminate type with upright stamens which produce an abundance of viable pollen and with only an aborted or rudimentary pistil. The other type of flower is the imperfect hermaphrodite with normal pistil and semi-reflexed stamens which yield aborted pollen.

Counts were made of the number of flowers in some 80 different clusters on vines growing in the mountains and in the Botanic Garden at Sapporo. These counts showed that clusters of male flowers contain more flowers than do clusters with imperfect hermaphrodite flowers. No germination of pollen was obtained from the shrivelled pollen of the semi-reflexed stamens. The pollen from the erect stamens of male flowers was highly viable on artificial media.

It may be reported that in the various studies on the longevity of grape pollen of numerous varieties thus tested only the pollen of Alicante, a greenhouse *vinifera* variety, gave 4 per cent germination just one year after it was collected. This pollen was kept stored in a dessicator using calcium chloride as the drying agent. A 10 per cent sucrose solution in redistilled water was used for germination.

#### ACKNOWLEDGMENT

In these experiments the author wishes to acknowledge his indebtedness in use of materials and in eager assistance to the following gentlemen: Professor Y. Hoshino of Hokkaido Imperial University, Japan; Professor F. Bioletti and Mr. L. O. Bonnet at Viticultural Department University Farm, Davis, California; Mr. H. Kawakami, Jr., of Niigata, and Mr. T. Tanaka and Mr. Niizuma of Hokkaido, Japan.





All are fruits of the grape Madelaine Angevine.

1. Poorly filled clusters, the best obtained by self-pollination.
2. Clusters obtained with stored pollen of White Corinth.
3. Clusters obtained with stored pollen of wild grape *Vitis Coignetiac*.
4. Well-filled clusters obtained when there is proper cross-pollination. The pollen used in this case was fresh from Precoce de Malingre.



## SOME STERILE AND FERTILE PLANT HYBRIDS

N. E. HANSEN

*Agricultural College of South Dakota*

During the past 31 years, devoted to the improvement of fruits, the writer has originated many hybrids. A brief mention will suffice at this time. Some species yield fertile hybrids with certain species, and sterile hybrids with other species. The reason for this is not yet clear. But homozygous material in general gives better results than heterozygous material as in complex hybrids, where apparently the chromosome structure becomes too complex or inharmonious.

*Prunus Besseyi*  $\times$  *P. triflora*. Highly fertile. Bears on one-year-old shoots from the bud in nursery. Example: Opata, Sapa. These are widely grown in our prairie Northwest. The South Dakota Sand Cherry is dominant in hardiness and in habit of bearing; the Japanese plum transmits quality and large size of fruit.

*Prunus Besseyi*  $\times$  *P. Simonii*. Not hardy. Fruits sparingly. Tokeya, the only one named, was discarded.

*Prunus Besseyi*  $\times$  *P. americana*. The named varieties, Sansoto and Cheresoto, are highly fertile. The quality not up to Opata and Sapa. This is to be expected as neither parent possesses high quality.

*Prunus Besseyi*  $\times$  *P. Pissardi* (*P. cerasifera purpurea*). Fruits of no value and sparingly produced. But as ornamental shrubs with red leaves the varieties Cistena and Stanapa are popular in western gardens.

*Prunus Besseyi*  $\times$  *P. Armeniaca*. Very shy bearer. Not of thrifty growth, rather slender.

*Prunus Besseyi*  $\times$  *P. avium*. Growth very dwarf and plants soon perished.

*Prunus Besseyi*  $\times$  *P. Persica*. Kamdesa, my hybrid of the Sand Cherry of South Dakota with the peach, is of special interest. It has shown fruit only once in its history. The flowers instead of one pistil, have two to six pistils. The pollen is sterile. In this connection should be recalled the sterile peach  $\times$  plum hybrids that have appeared in various places.

*Prunus Besseyi*,  $F_2$  hybrids. All highly fertile, when as in my Tom Thumb and Oka, the pedigree is evidently three-fourths Sand Cherry and one-fourth Japanese plum (*P. triflora*).

One of my hybrids, combining four species, *P. Besseyi*, *P. Simonii*, *P. americana* and *P. triflora*, is nearly sterile. Selfing with plums I find very difficult.

*Prunus americana*  $\times$  *P. Simonii*. My varieties Hanska and Kaga give choice fruit and are productive in mixed orchards. Inkpa, the sister variety,

is not productive. All have the excelled quality and fragrant firm-fleshed fruit of the *P. Simonii* of China.

*Prunus Simonii*  $\times$  *P. americana*. Tokata is larger in fruit and considered one of the very best in flavor. Needs good pollination to produce fruit. It is evident that *P. Simonii* gives high quality. Most hybrids must have good pollination in mixed orchards.

*Prunus triflora*  $\times$  *P. americana*. The hybrids are all highly fertile. The fruit of my Waneta is two inches in diameter. The tree is very strong and vigorous in growth. Also the two sister varieties, Kahinta and Tawena.

*Prunus americana*  $\times$  *P. triflora*. The many choice hybrids produced by others as well as myself are fertile and early in bearing and show clearly that these two species are mutually fertile.

*Prunus Americana*  $\times$  *P. domestica*. These two species combine either way with difficulty. They are not valuable and are generally sterile so far as noted in my experiments.

*Prunus nigra*  $\times$  *P. triflora*. The native plum of Manitoba, *Prunus nigra*, combines well with the Japanese plum and its hybrids. Examples, my Cree, Pembina and Ojibwa.

*Prunus nana*  $\times$  *Persica*. The Siberian almond always gives sterile hybrids with the peach but they are desirable ornamentals.

*Pyrus baccata*  $\times$  *P. Malus*. Thousands of hybrids have appeared since the Siberian crab-apple was brought to America. *Pyrus Malus* is considered by C. Koch to be a composite of six species, hence great variety is found in all the *P. Malus* hybrids. These hybrids are usually productive. When sterility is evident they are soon discarded. The best are of economic importance as a fruit for preserving. When too large they are not of importance, as they are too large for a crab-apple and too small for an apple. Some of the best crabs, like my Dolgo, are direct importations from Russia. The pedigree is not known. My Alexis crab which is very similar in every respect to Dolgo, was grown from seed of *Pyrus baccata* received from the Botanical Garden at Leningrad.

*Pyrus Malus*  $\times$  *P. baccata*. My best so far is Maga crab, a hybrid of McIntosh apple with the Virginia crab (a hybrid crab). My Olga crab (Duchess of Oldenburg apple  $\times$  *P. baccata cerasifera*) is highly fertile.

My Hopa red-flowered crab (*P. Malus Niedzwetzkyana*  $\times$  *baccata*) is highly fertile. The fruit is small, but the tree is very ornamental when in bloom.

The longer I work with *Pyrus baccata* the more I am convinced that pure selection work should be done with this species. Apparently the best form is the most northern type so far available. The Nertchinsk seedlings from the Amur River region of eastern Siberia are wonderfully productive trees. Since my best South Dakota sand cherries this year are one inch in diameter, the product of selection through several plant generations, I believe that the pure Siberian crab, *Pyrus baccata*, can be developed to full apple size by



selection only, without the hybridization with the cultivated apple, *Pyrus Malus*, which sometimes gives sterility or lack of winter hardiness.

*Native American Apple.* The wild west American apples *Pyrus Soulandi* and *P. ioensis* combine well with the cultivated apple, *P. Malus*. (Note my Kola, Zapta, Tipi and Shoko.) But the work is not finished as the wild crab acerbity is yet too much in evidence. My Anoka apple is very popular as it is of good quality and bears in one or two years after transplanting one year budded trees on one-year-old wood. It is from seed of the hybrid wild crab Mercer topgrafted on Duchess of Oldenburg. The tree is a remarkable annual bearer and remains semi-dwarf from its heavy bearing.

*The Future Program for the Apple.* If it is true that six species are in the ancestry of the cultivated apple, it would be an extremely difficult piece of work to reduce the apple to the homozygous condition, which is so desirable for  $F_1$  combinations. Homozygosity is the ideal of the apple breeder, but it appears very difficult to secure this judging from the experiments so far in several states. Many valuable varieties have been obtained by the use of entirely heterozygous material, but it would be well to go further and determine the results of using purely homozygous material.

*Hybrid Alfalfas.* Alfalfa is an old Arabic word meaning the best fodder. More and more alfalfa is becoming the great essential to successful farming over a large area of the United States. In Asia and southern Europe wherever the common blue-flowered alfalfa, *Medicago sativa*, and the yellow-flowered alfalfa, *Medicago falcata*, grow near together the hybridization takes place freely. Botanists call these hybrid forms *M. media* and the farmer calls them "Sand Lucerns." These hybrids are very numerous and consist of all sorts of mixtures in varying proportions of the yellow and blue alfalfa. As a class they are very productive and more desirable than either parent in vigor and productiveness.

Since *Medicago falcata* is very widely distributed in Europe and Asia, ranging in Asia from India north to above the Arctic Circle in north-eastern Siberia, the plant varies greatly in its ability to resist cold, hence it follows that hardiness of this hybrid alfalfa must depend largely on the region from which it comes. Coming from the mild region of southern Europe it could not be expected to be as hardy as if it came from drier and more severe climates. Hence while nature has indicated in the Sand Lucernes a method of increasing the vigor of alfalfa by hybridization, we do not know, that any one combination is the best one that it is possible to make.

The strongest and best of these hybrid alfalfas is the one I brought from Russia in 1906 and named the Cossack. The small spoonful of seed which I brought over in 1906 has been developed in the hands of many farmers so that the 1916 crop in the western part of South Dakota was fully 1000 bushels and since then the acreage has steadily increased.

These hybrid alfalfas as a class are superior to either parents in vigor and productiveness. I have originated many varieties by alternate machine transplanting of one-year-old plants of two varieties as first noted in South

Dakota Bulletin, 159, April, 1915. But pressure of other work has prevented their further development and propagation. South Dakota No. 1 and South Dakota No. 2 are the only two of this series of hybrids that have been distributed. The chief trouble is that the variable variegation in the color of the flowers makes it impossible to identify them. So their sale is entirely a matter of good faith. This need of a definite trademark led me to work for a white-flowered alfalfa.

This has been done in my white-flowered alfalfa and later in this present year, 1926, in my Hansen white-seed alfalfa, which has both white flowers and white seeds. It is the only alfalfa ever introduced that has a distinguishing characteristic so that it does not need certification or affidavits as to genuineness.

### CONCLUSION

For further light in this matter it will be necessary to make all possible combinations of species. We will then know better what species are mutually congenial. To cross heterozygous with heterozygous parents is like aiming in the dark. But as Nature produces them in vast numbers, some good results are bound to be obtained. I favor using homozygous material as much as possible as greater hybrid vigor is obtained.

# SELF AND CROSS-STERILITY IN THE JAPANESE PEAR

AKIO KIKUCHI

*Tottori Agricultural College, Tottori, Japan*

The origin of the cultivated pear in Japan is unknown. It may have its beginning in the wild species of the country or it may have been brought from China or Korea. There is no evidence to show when its improvement began, nor when the fruit reached the size, color and quality of what is considered a good variety today.

It is supposed, however, that as early as the eight century of the Christian era or even in a more remote period the people cultivated the pear or had some knowledge of its cultivation. We have a quotation from a book entitled "Nihon-Shoki," which reads as follows: "The Government advised the people to plant pear trees, chestnut trees, etc., to increase the food supplies of the nation." This is believed to be a publication of the eighth century.

Although the history of the cultivated pear dates back many years, its commercial importance is so recent that it only became one of the chief crops of orcharding in the latter part of the last century.

The range of the commercial adaptability of the Japanese pear is very wide, so wide, indeed, that it is grown successfully in commercial quantity in almost every fruit-growing section of the country except the regions where tropical fruits are produced. Botanically, the present varieties of the Japanese pear belong to *Pyrus serotina* Rehder, the species which is native to the middle and southern parts of China and the southern part of Korea.

We cannot find the true wild form of this species in Japan. More than one hundred varieties of this fruit may be found in Japan, but the varieties commercially grown are not more than fifteen in number, most of which have originated as chance seedlings during the past forty years.

The author intended to improve the quality of the fruit of the Japanese pear, which is generally believed to be very poor in comparison with that of the European varieties. The breeding work was begun in 1914 in connection with some studies on the sterility in named varieties of the Japanese pear. In all, 1,300 seedlings have been raised during the past ten years and 90 per cent of those have reached bearing age today. Studies on their sterility were also carried on with these plants.

## THE MATERIALS USED AND THE GENERAL PLAN OF THE INVESTIGATION

Twenty-four named varieties were used for pollination experiments, selfing and crossing, to study self- and cross-compatibility of each variety.

The name of the variety as follows: Chojuro, Nijisseiki, Kozo, Wase-Aka, Sekiriu, Imamura-Aki, Shinchiu, Okusankichi, Akappo, Doitsu, Taihei, Hakuteiriu, Taihaku, Sekai-Ichi, Meigetsu, Wase-Chojuro, Kinchaku, Amanogawa, Ohiromaru, Okoga, Wase-Kozo, Imamura-Natsu, Ichihara and Hakata-Awo.

The following plants were used in this work:

Imamura-Aki $\times$ Chojuro .....	23	plants
Chojuro $\times$ Akappo and Akappo $\times$ Chojuro.....	49	"
Chojuro $\times$ Nijisseiki and Nijisseiki $\times$ Chojuro.....	26	"
Chojuro $\times$ Kozo .....	17	"
Taihaku $\times$ Nijisseiki and Nijisseiki $\times$ Taihaku.....	12	"
Chojuro $\times$ Meigetsu and Meigetsu $\times$ Chojuro.....	9	"
Chojuro $\times$ Doitsu .....	9	"
Taihaku $\times$ Chojuro and Chojuro $\times$ Taihaku.....	12	"

These plants were tested for self-sterility, and for cross-sterility with both parent plants, and inter-crossings were also made among the progenies from the same parents.

## EXPERIMENTAL RESULTS WITH NAMED VARIETIES

Experimental results in self-pollination in twenty-four named varieties are shown in TABLE 1. This work was carried on in five orchards in three different prefectures. The situation of the orchards for which data are presented in TABLE 1 is indicated as follows:

Tokyo: The orchard of the Tokyo Horticultural School.

Kanagawa (a): The Ninomiya Horticultural Branch Station, The Kanagawa Agricultural Experiment Station, The Kanagawa Prefecture.

Kanagawa (b): The former experiment station of the Kanagawa Prefecture Hodogaya, near Yokohama.

Tottori (a): A grower's orchard, near the Tottori Agricultural College.

Tottori (b): The orchard of the Tottori Agricultural College.

In looking over TABLE 1 it is to be noted that Chojuro, Kozo, Shinchiu, Taihei, and Hakata-Awo are self-sterile varieties of remarkable stability. On the other hand, Nijisseiki, Okusankichi, Kinchaku, Ohiromaru and Okoga have proved to be not so stable as in other varieties in respect to the self-sterility. Chojuro, Nijisseiki and Okusankichi are leading varieties of today in Japan. This experimental results have shown some coincidence with the observation of practical growers with respect to the point of view.

Although there is some fluctuation in the stability of self-sterility, the other varieties, as shown in TABLE 1, have proved to be practically self-sterile in this experiment. Numerous crosses were made among twenty-three named varieties and those experimental results are shown in TABLE 3, in which numerical data are omitted.

There are four cases of the cross-sterility among 106 matings as shown in TABLE 3. Wase-Aka  $\times$  Taihaku and Meigetsu  $\times$  Ichihara gave



cross-sterility, and reciprocals showed also the same results respectively. When above four varieties were crossed with other varieties, results proved to be perfectly fertile. Numerical data for this experiment are shown in TABLES 2 and 4. The germination test of the pollen of all varieties was made with 20 per cent cane sugar solution. All varieties were proved to have perfect germinability.

### EXPERIMENTAL RESULTS WITH $F_1$ PLANTS

A. Experiments of self-pollination were conducted in 1925 and 1926.

1. Chojuro  $\times$  Nijisseiki and its reciprocal cross. Among 37 plants of the same progenies the following seven individuals set fruit in self-pollination in one season or two.

Tree No.	% set in 1925	% set in 1926
68	0	40.00
201	26.67	6.67
205	20.00	46.67
206	0	20.00
208	20.00	0
751	.....	6.67
762	0	6.67

Other individuals set no fruit in the both seasons.

2. Chojuro  $\times$  Akappo and its reciprocal cross. Among 47 individuals No. 19 gave 13.33 per cent set in 1924, but no fruit in 1926. Other 46 plants gave no fruit in both seasons.

3. Imamura-Aki  $\times$  Chojuro. Among 20 plants from the same parents, 9 individuals set no fruit in both seasons, and following 11 gave fruit set.

Tree No.	% set in 1925	% set in 1926
422	13.33	0
423	6.67	8.33
425	4.17	0
426	0	16.67
427	3.33	0
429	12.50	0
432	6.67	.....
434	10.00	0
435	13.33	4.17
439	26.67	0

4. Nijisseiki  $\times$  Akappo and its reciprocal cross. The testing was made once in 1925. Among 30 plants, No. 87 gave 12.50 per cent set, and all other individuals set no fruit.

5. Chojuro  $\times$  Kozo. Eight individuals were used for this experiment. three from those gave fruit set in 1926.

Tree No.	% set in 1925	% set in 1926
409	0	40.00
411	0	20.00
412	0	6.67

6. Chojuro  $\times$  Talhaku and its reciprocal cross. All individuals gave no fruit in 1926. The test was not made in 1925.

B. Cross-sterility occurred between  $F_1$  plants and their parents. Crossing experiments were made to study the occurrence of cross-sterility between  $F_1$  plants and their parents. Experimental results are shown in the TABLE 5.

It will be noted in this table that there is no occurrence of cross-sterility in mating between  $F_1$  plants and plants of pistil parent.

The cross-sterility will be seen always in matings of  $F_1$  plants between their pollen parent.

C. Cross-sterility occurred in inter-crossing between  $F_1$  plants from the same parents.

1. Imamura-Aki  $\times$  Chojuro. Among 56 matings between these progenies, following matings gave cross-sterility.

423 $\times$ 425	425 $\times$ 423	425 $\times$ 433	433 $\times$ 425
427 $\times$ 428	428 $\times$ 427	435 $\times$ 436	436 $\times$ 435
*430 $\times$ 429	...	*423 $\times$ 433	...

When 429 was crossed with pollen of 430, it set 11.86 per cent (two fruits out of 17 flowers). The occurrence of cross-sterility amounts to 17.86 per cent (10 from 56 matings).

2. Chojuro  $\times$  Kozo. Inter-crossings were made among only eight individuals of these progenies, but occurrence of cross-sterility was so remarkable as is shown in the following tabulation:

409 $\times$ 410	410 $\times$ 409	410 $\times$ 411	411 $\times$ 410
412 $\times$ 413	413 $\times$ 412	413 $\times$ 414	414 $\times$ 413
409 $\times$ 416	416 $\times$ 409	411 $\times$ 412	...
415 $\times$ 416	...	*409 $\times$ 412	...
*409 $\times$ 413	...	*410 $\times$ 415	...

The crosses 411  $\times$  412 and 415  $\times$  416 set no fruit altogether, but the reciprocal crossing gave 6.25 per cent and 14.29 per cent (1 fruit out of 16 flowers and 2 out of 14) respectively.

The occurrence of cross-sterility amounts to 57.69 per cent in this case (26 out of 56 matings).

3. Chojuro  $\times$  Nijisseiki and its reciprocal cross. Sixteen individuals of these progenies were used for this testing. Matings which were cross-sterile are as follows:

65 $\times$ 67	67 $\times$ 65	67 $\times$ 206	206 $\times$ 67	764 $\times$ 772	772 $\times$ 764
65 $\times$ 202	202 $\times$ 65	762 $\times$ 751	...	786 $\times$ 762	...
*68 $\times$ 764	...	*205 $\times$ 208	...	...	...

Reciprocal crossings of 762  $\times$  751 and 786  $\times$  762 gave 37.50 per cent and 47.06 per cent (6 fruits out of 16 flowers and 8 out of 17) respectively. Cross-sterility occurrence amounts to 20.69 per cent (12 out of 46 matings).

4. Chojuro  $\times$  Akappo and its reciprocal cross. Plant No. 34 was crossed with pollen of 42, 49 and 51. No fruit set in these matings. Crossings were made also on 40 with pollen of 33, 36, 41 and 42. Pollen of 41 set no fruit, but other three proved to be fertile with 40. The occurrence of the cross-sterility in inter-crossing between these progenies would perhaps be more if further studies were made.

\* The reciprocal crossings not tested.

\* Reciprocal crossings not tested.

\* Reciprocal crossings not tested.

TABLE 1  
EXPERIMENTAL RESULTS IN SELF-POLLINATION IN 23 VARIETIES

VARIETY	SEASON	ORCHARD	NUMBER FLOWERS USED	NUMBER SET	PER CENT SET	AVERAGE
Chojuro	1916	Tokyo	63	0	0.00	.06
Chojuro	1916	Tokyo	184	0	0.00	
Chojuro	1916	Tokyo	114	0	0.00	
Chojuro	1916	Tokyo	96	0	0.00	
Chojuro	1916	Tokyo	150	0	0.00	
Chojuro	1917	Tokyo	357	0	0.00	
Chojuro	1917	Kanagawa (A)	110	0	0.00	
Chojuro	1918	Kanagawa (A)	98	0	0.00	
Chojuro	1919	Kanagawa (A)	146	1	0.69	
Chojuro	1924	Tottori (A)	20	0	0.00	
Chojuro	1926	Tottori (B)	45	0	0.00	
Nijisseiki	1916	Tokyo	168	17	10.12	9.75
Nijisseiki	1917	Tokyo	212	45	21.23	
Nijisseiki	1917	Kanagawa (A)	56	20	35.71	
Nijisseiki	1918	Kanagawa (A)	98	2	2.04	
Nijisseiki	1918	Kanagawa (A)	97	4	4.12	
Nijisseiki	1919	Kanagawa (A)	50	2	4.00	
Nijisseiki	1919	Kanagawa (A)	100	6	6.00	
Nijisseiki	1924	Tottori	20	0	0.00	
Nijisseiki	1926	Tottori	44	2	4.55	
Kozo	1916	Tokyo	108	2	1.85	.51
Kozo	1917	Tokyo	176	3	1.71	
Kozo	1917	Kanagawa (B)	93	0	0.00	
Kozo	1917	Kanagawa (A)	140	0	0.00	
Kozo	1918	Kanagawa (A)	40	0	0.00	
Kozo	1926	Tottori (B)	45	0	0.00	
Wase-Aka	1916	Tokyo	252	7	2.78	
Wase-Aka	1917	Tokyo	204	0	0.00	
Wase-Aka	1917	Kanagawa (B)	162	4	2.47	
Wase-Aka	1917	Kanagawa (A)	12	0	0.00	
Wase-Aka	1918	Kanagawa (A)	96	0	0.00	2.02
Wase-Aka	1924	Tottori (A)	22	0	0.00	
Wase-Aka	1926	Tottori (B)	45	4	8.89	
Sekiriu	1917	Tokyo	252	2	0.79	
Sekiriu	1917	Kanagawa (B)	123	1	0.81	
Sekiriu	1917	Kanagawa (A)	65	0	0.00	
Sekiriu	1918	Kanagawa (A)	96	0	0.00	
Sekiriu	1924	Tottori (A)	22	0	0.00	
Sekiriu	1926	Tottori (B)	43	6	13.95	
Imamura-Aki	1916	Tokyo	162	3	1.85	1.58
Imamura-Aki	1917	Tokyo	243	11	4.53	
Imamura-Aki	1917	Kanagawa (A)	50	0	0.00	
Imamura-Aki	1918	Kanagawa (A)	101	0	0.00	
Imamura-Aki	1924	Tottori (A)	20	0	0.00	
Imamura-Aki	1926	Tottori (B)	44	0	0.00	
Shinchiu	1916	Tokyo	321	3	0.94	
Shinchiu	1917	Tokyo	224	1	0.45	
Shinchiu	1917	Kanagawa (A)	68	0	0.00	
Shinchiu	1918	Kanagawa (A)	95	0	0.00	
Shinchiu	1926	Tottori	45	1	2.22	0.72
Okusankichi	1917	Tokyo	360	89	24.72	
Okusankichi	1917	Kanagawa (A)	31	4	12.90	
Okusankichi	1918	Kanagawa (A)	60	1	1.67	
Okusankichi	1924	Tottori (A)	20	3	15.00	
Okusankichi	1926	Tottori (B)	9	5	55.55	
						21.97

TABLE 1—Continued  
EXPERIMENTAL RESULTS IN SELF-POLLINATION IN 23 VARIETIES

VARIETY	SEASON	ORCHARD	NUMBER FLOWERS USED	NUMBER SET	PER CENT SET	AVERAGE
Akappo.....	1916	Tokyo.....	273	0	0.00	2.52
Akappo.....	1917	Tokyo.....	163	10	6.14	
Akappo.....	1917	Kanagawa (A)...	40	1	2.50	
Akappo.....	1918	Kanagawa (A)...	66	1	1.52	
Akappo.....	1926	Tottori (B).....	41	1	2.44	
Doitsu.....	1916	Tokyo.....	297	5	1.68	5.19
Doitsu.....	1917	Tokyo.....	302	23	7.61	
Doitsu.....	1918	Kanagawa (A)...	38	0	0.00	
Doitsu.....	1924	Tottori (A).....	20	0	0.00	
Doitsu.....	1926	Tottori (B).....	30	5	16.67	
Taihei.....	1916	Tokyo.....	216	0	0.00	0.00
Taihei.....	1917	Tokyo.....	164	0	0.00	
Taihei.....	1918	Kanagawa (A)...	100	0	0.00	
Taihei.....	1926	Tottori (B).....	18	0	0.00	
Taihei.....	1926	Tottori (B).....	45	0	0.00	
Hakuteiriu.....	1916	Tokyo.....	114	0	0.00	12.34
Hakuteiriu.....	1917	Tokyo.....	215	58	26.98	
Hakuteiriu.....	1917	Kanagawa (A)...	44	7	15.91	
Hakuteiriu.....	1918	Kanagawa (A)...	93	6	6.45	
Taihaku.....	1916	Tokyo.....	198	1	0.51	
Taihaku.....	1917	Tokyo.....	308	11	3.57	4.33
Taihaku.....	1917	Kanagawa (A)...	63	0	0.00	
Taihaku.....	1918	Kanagawa (A)...	59	5	8.48	
Taihaku.....	1926	Tottori (B).....	33	3	9.09	
Sekai-Ichi.....	1917	Tokyo.....	128	10	7.81	
Sekai-Ichi.....	1918	Kanagawa (A)...	0	0	0.00	4.22
Sekai-Ichi.....	1924	Tottori (A).....	20	0	0.00	
Sekai-Ichi.....	1926	Tottori (B).....	44	4	9.09	
Meigetsu.....	1917	Kanagawa (B)...	93	4	4.30	
Meigetsu.....	1917	Kanagawa (A)...	33	0	0.00	
Meigetsu.....	1923	Kanagawa (A)...	20	0	0.00	5.60
Meigetsu.....	1924	Tottori (A).....	20	0	0.00	
Meigetsu.....	1926	Tottori (B).....	32	0	0.00	
Wase-Chojuro.....	1917	Kanagawa (A)...	93	2	2.15	
Wase-Chojuro.....	1918	Kanagawa (A)...	97	0	0.00	
Wase-Chojuro.....	1924	Tottori (A).....	20	0	0.00	1.65
Wase-Chojuro.....	1926	Tottori (B).....	45	2	4.44	
Kinchaku.....	1916	Tokyo.....	81	13	16.05	
Kinchaku.....	1917	Tokyo.....	163	10	6.14	
Amanogawa.....	1916	Tokyo.....	159	3	1.89	
Amanogawa.....	1917	Tokyo.....	451	14	3.10	2.79
Ohiromaru.....	1916	Tokyo.....	126	8	6.35	
Ohiromaru.....	1917	Tokyo.....	204	21	10.30	
Okoga.....	1917	Kanagawa (B)...	300	37	12.33	
Okoga.....	1918	Kanagawa (B)...	189	21	11.11	
						11.72



TABLE 1—Continued  
EXPERIMENTAL RESULTS IN SELF-POLLINATION IN 23 VARIETIES

VARIETY	SEASON	ORCHARD	NUMBER FLOWERS USED	NUMBER SET	PER CENT SET	AVERAGE
Wase-Kozo.....	1917	Kanagawa (B)...	72	1	1.39	3.19
Wase-Kozo.....	1918	Kanagawa (A)...	38	0	0.00	
Wase-Kozo.....	1926	Tottori (B).....	44	2	4.55	
Wase-Kozo.....	1926	Tottori (B).....	44	3	6.81	
Imamura-Natsu.....	1917	Kanagawa (A)...	82	2	2.44	3.59
Imamura-Natsu.....	1918	Kanagawa (A)...	91	0	0.00	
Imamura-Natsu.....	1926	Tottori (B).....	36	3	8.33	
Ichihara.....	1923	Kanagawa (B)...	24	0	0.00	2.38
Ichihara.....	1924	Tottori (A).....	20	0	0.00	
Ichihara.....	1926	Tottori (B).....	42	3	7.14	
Hakata-Awo.....	1924	Tottori (A).....	20	0	0.00	
Hakata-Awo.....	1926	Tottori (B).....	45	0	0.00	0.00

TABLE 2  
CROSS-STERILITY BETWEEN WASE-AKA AND TAIHAKU

VARIETY USED AS SEED PARENT	VARIETY USED AS POLLEN PARENT	SEASON	ORCHARD	NUMBER FLOWERS USED	NUMBER SET	PER CENT SET	AVER- AGE
Wase-Aka...	Taihaku...	1916	Tokyo.....	348	3	0.86	.72
Wase-Aka...	Taihaku...	1917	Kanagawa (B)	24	0	0.00	
Wase-Aka...	Taihaku...	1918	Kanagawa (B)	132	0	0.00	
Wase-Aka...	Taihaku...	1917	Kanagawa (A)	129	1	0.78	
Wase-Aka...	Taihaku...	1918	Kanagawa (A)	51	1	1.96	
Wase-Aka...	Ohiromaru..	1917	Kanagawa (B)	18	14	77.78	8.43
Wase-Aka...	Nijisseiki...	1917	Kanagawa (A)	60	43	71.67	
Wase-Aka...	Nijisseiki...	1918	Kanagawa (A)	25	25	100.00	
Wase-Aka...	Nijisseiki...	1918	Kanagawa (A)	46	38	82.61	
Taihaku.....	Wase-Aka...	1916	Tokyo.....	171	1	0.59	
Taihaku.....	Wase-Aka...	1916	Tokyo.....	87	0	0.00	
Taihaku.....	Wase-Aka...	1917	Kanagawa (B)	46	2	4.35	
Taihaku.....	Wase-Aka...	1918	Kanagawa (B)	137	11	6.36	
Taihaku.....	Wase-Aka...	1917	Kanagawa (A)	86	27	31.40	
Taihaku.....	Wase-Aka...	1918	Kanagawa (A)	50	4	8.00	
Taihaku.....	Nijisseiki...	1917	Kanagawa (A)	62	52	83.87	8.43
Taihaku.....	Nijisseiki...	1918	Kanagawa (A)	35	29	82.86	
Taihaku.....	Ichahara...	1918	Kanagawa (A)	29	9	31.03	
Taihaku.....	Meigetsu...	1919	Kanagawa (A)	20	19	95.00	



TABLE 4  
CROSS-STERILITY BETWEEN MEIGETSU AND ICHIHARA

VARIETY USED AS SEED PARENT	VARIETY USED AS POLLEN PARENT	SEASON	ORCHARD	NUMBER FLOWERS USED	NUMBER SET	PER CENT SET	AVER- AGE
Meigetsu....	Ichihara....	1918	Kanagawa (A) .	26	0	0.00	1.25
Meigetsu....	Ichihara....	1919	Kanagawa (A) .	58	0	0.00	
Meigetsu....	Ichihara....	1923	Kanagawa (A) .	40	2	5.00	
Meigetsu....	Ichihara....	1924	Tottori (A)....	40	0	0.00	
Meigetsu....	Imamura-Aki	1917	Kanagawa (A) .	41	38	92.67	4.29
Meigetsu....	Chojuro....	1923	Kanagawa (A) .	19	18	94.74	
Ichihara....	Meigetsu....	1919	Kanagawa (A) .	19	0	0.00	
Ichihara....	Meigetsu....	1919	Kanagawa (A) .	34	0	0.00	
Ichihara....	Meigetsu....	1923	Kanagawa (A) .	28	2	7.14	
Ichihara....	Meigetsu....	1924	Tottori (A)....	40	4	10.00	
Ichihara....	Imamura-Aki	1924	Tottori (A)....	30	20	66.67	
Ichihara....	Chojuro....	1923	Kanagawa (A) .	22	21	95.45	

TABLE 5  
RESULTS OF CROSSING  $F_1$  PLANTS AND THEIR PARENTS

	FERTILE WITH BOTH PARENTS	STERILE WITH POLLEN PARENT AND FERTILE WITH SEED PARENT	STERILE WITH SEED PARENT AND FERTILE WITH POLLEN PARENT	STERILE WITH BOTH PARENTS	TOTAL	PERCENT- AGE OF CROSS- STERILE WITH POLLEN PARENT
Chojuro x Akappo and its re- ciprocal.....	28	21	0	0	49	42.86
Chojuro x Nijisseiki and its reciprocal.....	19	7	0	0	26	26.92
Imamura-Aki x Chojuro.....	23	0	0	0	23	0.00
Chojuro x Kozo.....	11	6	0	0	17	35.30
Taihaku x Nijisseiki and its reciprocal.....	6	6	0	0	12	50.00
Chojuro x Meigetsu and its reciprocal.....	9	0	0	0	9	0.00
Chojuro x Doitsu.....	9	0	0	0	9	0.00
Taihaku x Chojuro and its reciprocal.....	12	0	0	0	12	0.00





# STERILITY CAUSED BY THE ASTER YELLOWS DISEASE

L. O. KUNKEL

Boyce Thompson Institute of Plant Research  
(WITH PLATE 15)

Most mosaic diseases of plants reduce the yield of fruits and seeds. But the seeds from plants having these diseases are usually viable. Since in most cases mosaic is not transmitted through seeds, it was perhaps to be expected that it would not greatly effect their viability. But even when mosaic is transmitted through seeds, as is true in the case of a number of different leguminous plants, it does not prevent germination. It is rather surprising, therefore, that "yellows diseases," a group of plant maladies closely related to mosaic, cause sterility.

Smith\* showed many years ago that very few of the pits from yellowed peach trees will germinate. The aster yellows disease which attacks the China aster and many other plants also causes sterility.

Aster yellows is an infectious chlorosis transmitted by the leaf-hopper *Cicadula sexnotata* Fall. It is not transmitted by several other sucking insects that feed on the aster. The agent causing yellows is unknown. Before the carrier-leafhopper can spread the disease it must feed on a yellowed plant. It is interesting that the leafhopper cannot transmit yellows immediately after feeding on diseased plants. An interval of at least ten days must elapse between the time when the insect first feeds on a yellowed plant and the time when it first becomes inoculative. This interval, which is known as the incubation period of the virus in the insect, suggests a development of the causal agent in the body of the leafhopper. In this and in several other ways the disease is similar to malaria which is transmitted to man by the mosquito *Anopheles*.

The effects of yellows on asters was studied during the past three years. Plants that contract yellows while they are young and in the rosette stage do not produce seeds. Hundreds of such aster plants grown in greenhouses and in garden plots were kept until they became mature but none produced seeds. This is unfortunate for it prevents the propagation of progenies from plants that show resistance to or tolerance for the disease. Three plants that became diseased while in the rosette stage are shown in B of PLATE 15. A healthy plant of the same age is also shown. The diseased asters have produced no flowers.

If plants are somewhat older at the time of infection they may blossom and produce seeds. The blossoms of such plants are, however, very different

---

\* Smith, E. F. Peach yellows: a preliminary report. U. S. Dept. Agr. Div. Bot. Bull. 9. 254. pp. 1888.

from those of healthy plants. Their floral parts are stunted and of a pale green color regardless of the color of the flowers of the variety to which the plant belongs. Diseased flowers never show the white, pink, red or purple colors characteristic of different varieties of asters. The green flowers usually produce seeds. These seeds may be dwarfed but are more often somewhat larger than the seeds of healthy plants. Large numbers of seeds from diseased flowers were planted under favorable conditions but none germinated.

The severity of yellows depends on the age of the plant at the time it becomes infected. Three plants showing the disease in different degrees of severity are shown in *C* of PLATE 15. The plant at the right became diseased while young; that in the middle, somewhat later; and that on the left, as it was nearing maturity. All three plants have green flowers and will produce sterile seeds.

In some cases one side or only a few branches of a plant may be diseased at the time it blossoms. The healthy parts of such plants bear normal flowers and produce viable seeds. But the seeds produced by diseased branches or on diseased portions of individual flower heads are sterile. A healthy flower and three partly diseased flowers are shown in *A* of PLATE 15.

Diseased plants do not always produce seeds even when they bear flowers. The flowers may develop into vegetative branches or may produce secondary flowers or even flower heads on their stigmas. The stigmas of secondary flowers may also develop into flowers or flower heads. It is not uncommon, therefore, to find diseased flowers borne in short chains. This abnormal development is associated with yellows on aster, phlox, mignonette and many other plants.

Aster yellows furnishes an excellent example of sterility due to a transmissible disease. When virus-bearing individuals of *Cicadula sexnotata* feed on healthy aster plants they transmit some agent which prevents the plants from producing normal flowers or viable seeds. This kind of sterility is doubtless associated with several of the different yellows diseases that attack plants.

#### EXPLANATION OF PLATE 15

##### ASTER YELLOWS

- A. A healthy flower and three flowers partly diseased with aster yellows.
- B. A healthy plant and three diseased plants.
- C. Three plants showing the aster yellows in different degrees of severity.



KUNKEL: ASTER YELLOWS





## POLLEN-TUBE BEHAVIOR WITH REFERENCE TO STERILITY IN DATURA\*

J. T. BUCHHOLZ and  
*University of Texas*

A. F. BLAKESLEE  
*Carnegie Institution of Washington  
Station for Experimental Evolution*

(WITH PLATES 16-18)

*Datura Stramonium* is highly self-fertile and is at least partially compatible when crossed with its extra-chromosomal mutants. The phenomenon of self-incompatibility, or cross-incompatibility between certain strains, such as is found in Orchids, *Corydalis cava*, strains of *Nicotiana*, etc., is not known for *Datura*. We have found no difficulty in making inter-specific crosses within the *Stramonium* group of this genus, whose members have erect capsules. However, the *Datura* group with nodding capsules, embracing such species as *D. innoxia*, *D. metel* and *D. meteloides*, have not been successfully crossed with any of the forms with erect capsules.

Our studies on the sterility between species and races of *Datura* are therefore based on a study of the crosses between *D. Stramonium* and *D. meteloides* and between *D. Stramonium* and some of its partially sterile extra-chromosomal mutants.

It was our purpose to test the effects, as manifested in pollen-tube growth, of an attempted cross between species which have never been successfully crossed and are therefore known to be fully incompatible. We did this in order to learn whether the incompatibility found here is due to a slowness of pollen-tube growth, or to other causes. We were also seeking information as to any definite abnormal phenomena which might be obvious in the pollen tubes themselves, when these are growing where there is a strong degree of cross-incompatibility, since a special technique for *Datura* pollen tubes had already been perfected in connection with our genetic studies.

### TECHNIQUE

The technique used in a study of pollen tubes is one of dissection. The flowers to be studied are prepared by emasculation of the stamens and are then allowed to mature until the stigmas are fully receptive, which is usually a few hours after the pollen in flowers of the same age begins to shed from the anthers. The flowers may be pollinated while attached to the plant in the garden, or removed to the laboratory with a portion of the branch, and kept in a glass of water. The latter procedure is necessary where a close control of the temperature during pollen-tube growth is desired.

The flowers are pollinated at some convenient time after the stigmas are receptive, and after suitable intervals such as 6, 12 or 15 hours the styles are

---

\* This cooperative investigation was made possible by grants from the Joseph Henry Fund of the National Academy of Science. Miss Mae Sensing served as laboratory assistant for a part of this work.

plucked off, scalded for about 2 minutes in hot water (about 70° C.) and placed in a killing fluid of 6 per cent formalin in 50 per cent alcohol. Here they may be kept for several hours or even several days until a convenient time for dissection.

The dissections should be made under water, in a petri dish and under a Greenough binocular dissecting microscope with about 18× magnification. Strong direct illumination of the object is desirable, accompanied by some transmitted light; a combination of these two which will make details most visible with a low magnification. The cortical portion of the style is the part to be removed and discarded. This is done by using forceps in one hand, and a fine, bent dissecting needle in the other.

The portion to be obtained by the dissection method is the central strand of conducting tissue (*Leitgewebe*), which is usually made up of a solid strand of greatly elongated spindle-shaped cells. The effect of the scalding is to render this strand of tissue more gelatinous and thus more easily separated by dissection from the cortex of the style. A young flower which has been pollinated before it is fully matured is very difficult to dissect and a flower pollinated on the previous day and fully mature when pollinated, is relatively easy to dissect. Flowers gathered in the garden on the day after they open afford good practice material for the beginner in this technique.

In dissecting, one should begin by making a pair of longitudinal scratches or slits down each side of the style with the point of the needle. A very fine No. 10 sewing needle, mounted on a wooden handle and with a short bent point 2 mm. long (made by heating) is satisfactory. The slits should extend from the edges of the flattened portion of the stigma through the length of the style. This slitting may be done more quickly just before the scalding as follows:

A wooden block is made with a small groove at one end (Fig. 1). In this groove a fragment of razor blade is imbedded, adjusted to a depth which cuts the outer cells of the cortex but not the axial strand of conducting tissue. The index finger of the left hand is held over the style with gentle pressure while it is pulled through this groove. Each style is cut twice on opposite sides from a point past the middle out to the stigma region where the scratch should be made to terminate in the edges, not on the surface of the flat part of the cortex. The style is then reversed and the other end of it treated in this manner in order to complete the two slits throughout the length of the style.

The slits or scratches should not be made too deep. The needle mentioned above may then be inserted between the cortex and the central core of the tissue. One should be careful to separate the entire strand of conducting tissue from the inside of the cortex and pull the two portions apart until a lenticular opening is produced at this point. This opening is gradually lengthened in both directions as the cortex is stripped away from the other portion. The pulling should proceed only in one direction at a time. In separating these parts the needle may be made to pierce the strip of cortex

which is to be discarded, while the remaining portions are held with a delicate pair of forceps.

When the cortical strip has been removed from one side of the style the operation is repeated for the other side, and the central strand of conducting tissue must be handled with great care. The bent needle may be used as a scraper against the cortex if the core of conducting tissue is not easily separated. By practice, a skill may be acquired by which all of the conducting tissue may be removed in a single strand, and this strand is continuous with the stigmatic cells at the stigma region. The broadened stigma region, where the operation is most difficult, is usually removed last.

The stylar strand thus obtained may be returned to the watch glass of killing-preserving fluid for an indefinite period.

Before staining it should be placed into water for about 10 minutes. Analin red (magenta) has been used most successfully as a stain, in an aqueous 1 per cent solution, but the staining qualities are improved by the addition of a little light green stain which gives the mixture a very dark red color. Our practice has been to add a few drops of the light green stain from an alcoholic solution until the color is darkened to an almost purplish hue.

Staining requires several hours at least, depending upon the strength of the stain. The entire strand is picked out of the water with a needle and placed in the stain. After staining, the strand is dipped into water for an instant to remove the excess stain, then touched to a piece of blotting paper which absorbs the excess water, thus preventing dilution of the clearing fluid. Next, the stylar strand is placed in a watch glass in concentrated lactic acid where it should remain a half hour or more and be kept covered. The lactic acid serves as a clearing fluid and also as a mounting medium.

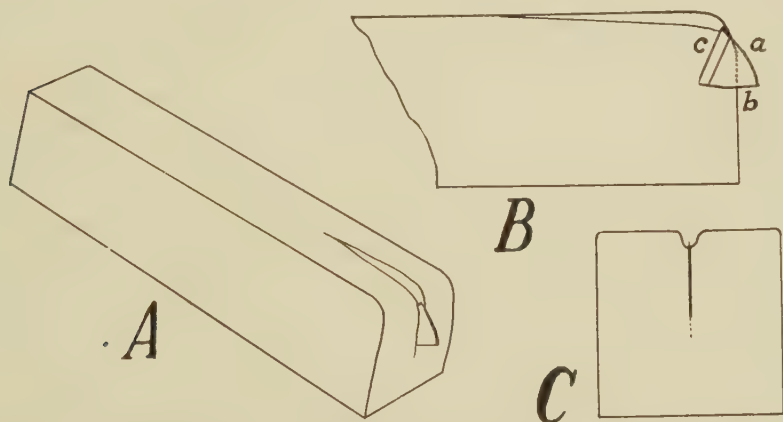


FIG. 1. Apparatus used in slitting cortex of styles, in making preparations of *Datura* pollen tubes. Block of wood with projecting point of razor blade within the groove is shown in a perspective view in A. B represents this apparatus in longitudinal section, showing the triangular fragment of razor blade in position with its cutting edge (c). Adjustment is made by tapping at a if the point projects too far, or at b if it is too low. C represents a transverse section through this block showing the point of the knife in the groove. Gentle pressure of the index finger holds the style in position during the operation.

In transferring the strand of tissue to a slide, it should be handled with a small camel's hair brush. It should be picked up by its large end and placed lengthwise on the slide, where it should be straightened out and made as free from twists as possible. A little lactic acid is added and a long cover glass is carefully placed over the strand, avoiding air bubbles. Then the strand is flattened out by gentle pressure applied to the top of the cover glass until the tissue has spread out as thin as desired. The dark red pollen tubes may now be observed as deeply stained thread-like structures imbedded in the lighter colored conducting tissue. The pressure will displace the pollen tubes slightly, but usually only laterally. They will maintain fairly well their relative distance between the stigma and the ovary.

These preparations make permanent mounts, good for several years if properly sealed at the edges. We have used Damar dissolved in xylol for this purpose applied with a small camel's hair brush. Usually we keep the preparations unsealed for a few weeks, press them out thinner and add more lactic acid if necessary before sealing. A double seal is desirable.

Where slides are to be sealed it is desirable to cut down the width of the cover glasses if necessary so that a margin of 2-3 mm. or more remains on all sides. The 35x62 mm. No. 2 cover glasses are excellent for this purpose when cut lengthwise into two strips with a piece of carborundum or a diamond. These are stronger and thicker covers than those of the smaller sizes and serve well in pressing out the tissues, but, of course, if oil immersion lenses are to be used it may be desirable to use No. 1 cover glasses.

The nuclear contents of the pollen tubes are not distinguishable by this method which stains the ends of the entire tube a very deep color. However, this technique has the advantage of showing in a single preparation all of the pollen-tube population of a single flower, and it also shows all, or nearly all, of the ungerminated pollen grains still adhering to the stigma, giving one an index to the proportions of germinated and ungerminated pollen.

From slides prepared in this manner it is possible to measure by means of a mechanical stage of a microscope the distance to which the foremost pollen tubes have penetrated, the place where most of them are found (the mode of their distribution), and to determine the grouping and distribution of the pollen tubes all along the style back to the stigma.

The pollen tubes are deeply stained only near the forward end. They all actually extend back to the stigma where each connects up with the empty shell (the exine) of the pollen grain from which it was derived. In making quantitative counts and measurements we have counted only the ends and measured the distance from these ends to an arbitrary point chosen a little below the surface of the stigma, a point which represents the average level of the pollen grains on the stigma. This point is usually about .7 mm. below the highest portion of the stigma in a prepared slide.

#### EXPERIMENTAL METHODS

In carrying out the observations and experiments which are reported here, about 10 cm. of the stem bearing the flowers together with several



leaves were cut from the plant under water and placed in bottles or tumblers. Castration was usually done before cutting, and the flowers were prepared and usually gathered early in the morning, at least before the anthers had shed pollen. The flowers which were used were fully mature, those which would have opened on the same day. The stamens for the pollen which was to be used were also gathered at this time and kept in covered watch glasses where the anthers dehisced during the day, and the pollinations were made in the evening (6-11 P. M.) of the same day. This practice yielded material which could be easily dissected, but we found that if the pollinations were made too early on the same day (for example before noon) the material was usually much more difficult to dissect. In the garden, the flowers open about 4-5 P. M., depending somewhat upon weather conditions.

A cave under the laboratory building of the Carnegie Institution of Washington at Cold Spring Harbor was found to be the most satisfactory place for a study of pollen-tube growth. This cave was constructed for the study of cave animals and usually maintains a relatively constant temperature around 18° C., a temperature and humidity at which excellent results in pollen-tube growth were always obtained with normal plants of *Datura Stramonium*, and in which nearly all of the pollen germinated.

The groups of flowers which had been collected during the morning were placed here and pollinated in the evening at some time after 7 P. M., then collected and killed after known intervals of time.

Pollinations were made with camel's hair brushes which had been previously sterilized and thoroughly dried out.

### NORMAL POLLEN-TUBE GROWTH

It will be necessary for us to describe here some of our results on a study of normal pollen tube growth in *Datura Stramonium*. For example, in the course of our studies a set of trials was made to determine the effect of time alone on pollen-tube growth. Does the rate of pollen-tube growth continue to be uniform as the tubes approach the ovary, or does the rate of pollen-tube growth change? Our earlier experiments had already convinced us that the temperature influences the pollen-tube growth profoundly, so that it was necessary for us to conduct these experiments in a place with a very uniform temperature, for which the cave mentioned above proved to be very satisfactory. On the shelf where the flowers of this experiment were kept, the temperature was 17.8° C.

In making measurements on slides obtained from the styles of the flowers used in this experiment, we found that several distances might be selected for this comparison. In TABLE 1, the measurements are given from a fixed point, .7 mm., from the top of the stigma (representing the average at which the pollen grains germinated on the curved surface of the stigma) to four such easily determined points which are as nearly comparable as possible under the slightly variable conditions where slightly different amounts of

TABLE 1  
POLLEN-TUBE GROWTH IN *Datura Stramonium*  
SELFS AND SIB CROSSES DURING VARIOUS TIME INTERVALS

TIME INTERVAL	DISTANCE REACHED BY LONGEST POLLEN TUBE MM.	DISTANCE REACHED BY 10TH POLLEN TUBE, MM.	DISTANCE REACHED BY 20TH POLLEN TUBE, MM.	DISTANCE OF APPROX- IMATE MODE, MM.	AVERAGE OF ALL MEASURE- MENTS	CORRECTED AVERAGE (x 1.06)
1 Hour B. ....						
Average. ....	0.34	0.30	0.29	0.28	0.30	0.32
2 Hours B. ....	3.2 2.9 2.5	2.9 2.3 2.0	2.6 2.1 1.8	2.2 1.7 1.5		
Average. ....	2.9	2.3	2.2	2.1	2.4	2.5
3 Hours B. ....	5.1 5.4	4.8 5.2	4.7 5.0	4.3 4.5		
A. ....	4.7 5.0	4.3 4.5	4.1 4.3	3.7 3.6		
Average. ....	5.1	4.7	4.5	4.0	4.6	4.9
6 Hours B. ....	14.1 13.8 13.6	13.4 13.6 12.3	13.1 13.4 12.1	11.6 12.2 10.9		
Average. ....	13.8	13.1	12.9	11.6	12.8	13.6
9 Hours B. ....	22.9 21.5 21.2	22.3 20.5 20.0	21.9 19.9 19.4	20.6 18.8 16.9		
A. ....	22.4 20.3	21.2 18.9	20.5 18.2	19.5 17.3		
Average. ....	21.7	20.6	20.0	18.6	20.2	21.4
12 Hours B. ....	33.8 30.4 30.2	31.5 28.4 29.2	31.1 28.1 28.4	28.8 25.8 24.4		
A. ....	29.2 28.8 27.3	26.3 27.5 26.4	26.7 27.0 26.0	25.6 25.8 25.0		
Average. ....	29.9	28.2	27.9	25.9	28.0	29.7
15 Hours B. ....	39.1 38.1 44.0	37.8 36.8 42.9	37.1 36.3 42.2	35.7 34.5 40.3		
A. ....	33.5 32.8	31.7 31.7	30.8 31.2	27.9 30.2		
Average. ....	37.5	36.2	35.5	33.7	35.7	37.8
18 Hours B. ....	45.0 48.8 46.2	43.9 47.0 45.7	43.2 46.4 44.8	40.6 43.6 42.5		
A. ....	45.0 45.9 43.1	43.2 44.8 41.8	43.8 44.3 41.1	42.3 42.3 39.7		
Average. ....	45.7	44.4	43.9	41.8	44.0	46.6
20 Hours B. ....	52.1 52.2 52.8	50.2 50.5 50.6	49.6 49.6 49.2	49.2 48.6 47.8		
A. ....	54.3	53.8	52.9	50.9		
Average. ....	52.8	51.3	50.3	49.1	50.9	....

TIME INTERVAL	DISTANCE REACHED BY LONGEST POLLEN TUBE MM.	DISTANCE REACHED BY 10TH POLLEN TUBE, MM.	DISTANCE REACHED BY 20TH POLLEN TUBE, MM.	DISTANCE OF APPROX- IMATE MODE, MM.	AVERAGE OF ALL MEASURE- MENTS	CORRECTED AVERAGE (x 1.06)
21 Hours A.....	59.0 48.7 52.9	52.6 46.5 51.8	51.2 45.7 51.2	49.5 43.7 49.4		
Average.....	53.5	50.3	49.4	47.5	50.2	....
20½ Hours (Average of A and B).....					50.6	53.6
22 Hours A.....	58.5 54.4 54.6 57.2	56.4 52.1 52.6 50.6	56.2 51.5 52.2 49.6	54.1 50.7 46.8 45.6		
Average.....	56.2	52.9	52.4	49.3	52.7	55.9

pollen had been applied to the different stigmas. The four sets of measurements include: (1) the distance to the end of the foremost or longest pollen tube; (2) the distance to the tenth pollen tube; (3) the distance to the 20th pollen tube; (4) the distance to the apparent mode. The latter was not the mode as determined by a critical count of an entire style, but rather to this point as observed by inspection, the place where the ends of the pollen tubes seemed to be most numerous. An examination of PLATE 17 at C will illustrate the appearance of the advancing front of a pollen-tube population and can give an idea of the clearness with which the pollen tubes may be observed with our technique. Thus each flower was used for four measurements and all of the different flowers were averaged with each other as a means of giving a general average for the distance of pollen-tube penetration after various intervals. We are presenting these tables completely in order to give readers an opportunity to note the amount of variation in these measurements.

At a constant temperature the rate of pollen-tube growth is practically the same in the various regions of the style. For a temperature of 17.8° C. this rate is 2.7 mm. per hour. Whether this rate changes at different times of the year or at different periods in the vegetative-reproductive cycle of any plant, we have not determined thus far. The variability found in TABLE 1 may be taken to suggest that it does.

A correction must be added if the figures of TABLE 1 are to be used as a basis for the estimate of this absolute distance of pollen tube penetration. This comes from the fact that the styles shrink considerably in the killing-preserving fluid. This shrinkage amounts to almost 6 per cent and the figures of TABLE 1 were therefore multiplied by the factor 1.06 in order to give the corrected distance in millimeters.

These experiments were based on normal sib crosses in homozygous material. Self-pollinations gave the same results as sib crosses. The plants of these stocks of *Datura Stramonium* were as nearly homozygous as it is

possible to produce them. They were derived as inbred strains through a haploid plant, and were therefore, barring new mutations, as nearly identical as cuttings all derived from a single plant.

There were only slight differences in the sizes of the flowers, due to differences in their growth vigor. It was necessary to use so many flowers that these could not be graded as closely for size as might be desired, but extremes in sizes were avoided.

Sib crosses of *Datura meteloides* were also made in the same manner in order to obtain a measure of the pollen-tube growth in this species. The flowers of this species are more than 17 cm. long at the time of opening. Dissection is much more difficult in *Datura meteloides* as the styles are usually about 165 mm. long. These flowers were also kept at 17.8° C. and gathered at various intervals. The distances of penetration of the longest pollen tubes were plotted individually for each flower in the graph of Fig. 3. These measurements show more variability than do those for *D. Stramonium*, but this difference may be partly due to the fact that the loci plotted here are not based on the combined averages of several flowers, as in the case of *D. Stramonium*. The maxima of each group also approach a straight line curve. In *D. meteloides* at 17.8° C. the style could be completely traversed by the pollen tubes in about 65 hours.

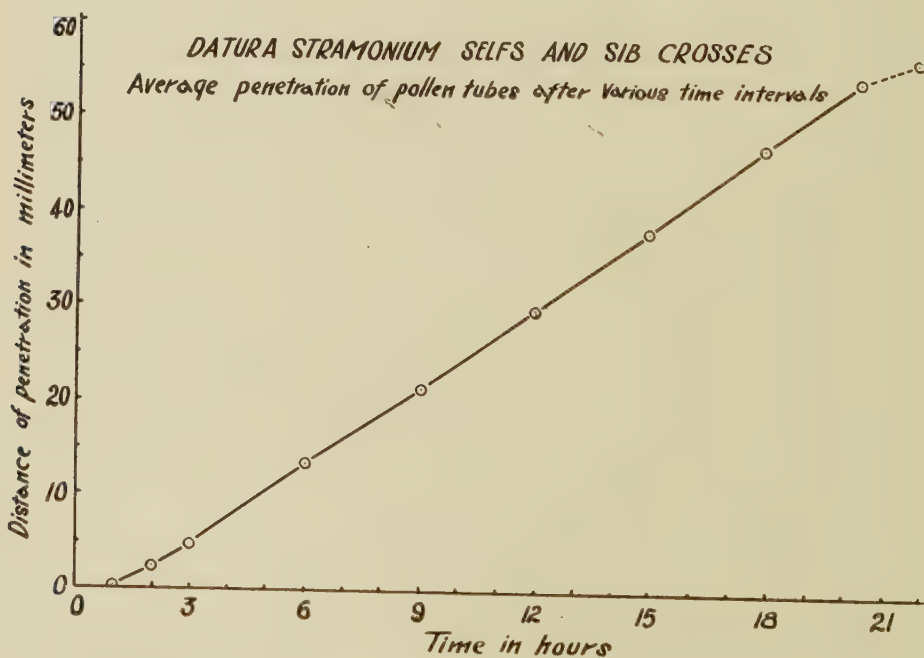


FIG. 2. Graph showing the relation of time to distance of penetration in the normal pollen-tube growth in *Datura Stramonium*.

Contrary to our expectation from the work of East and Park (3), there is neither an acceleration nor a retardation.



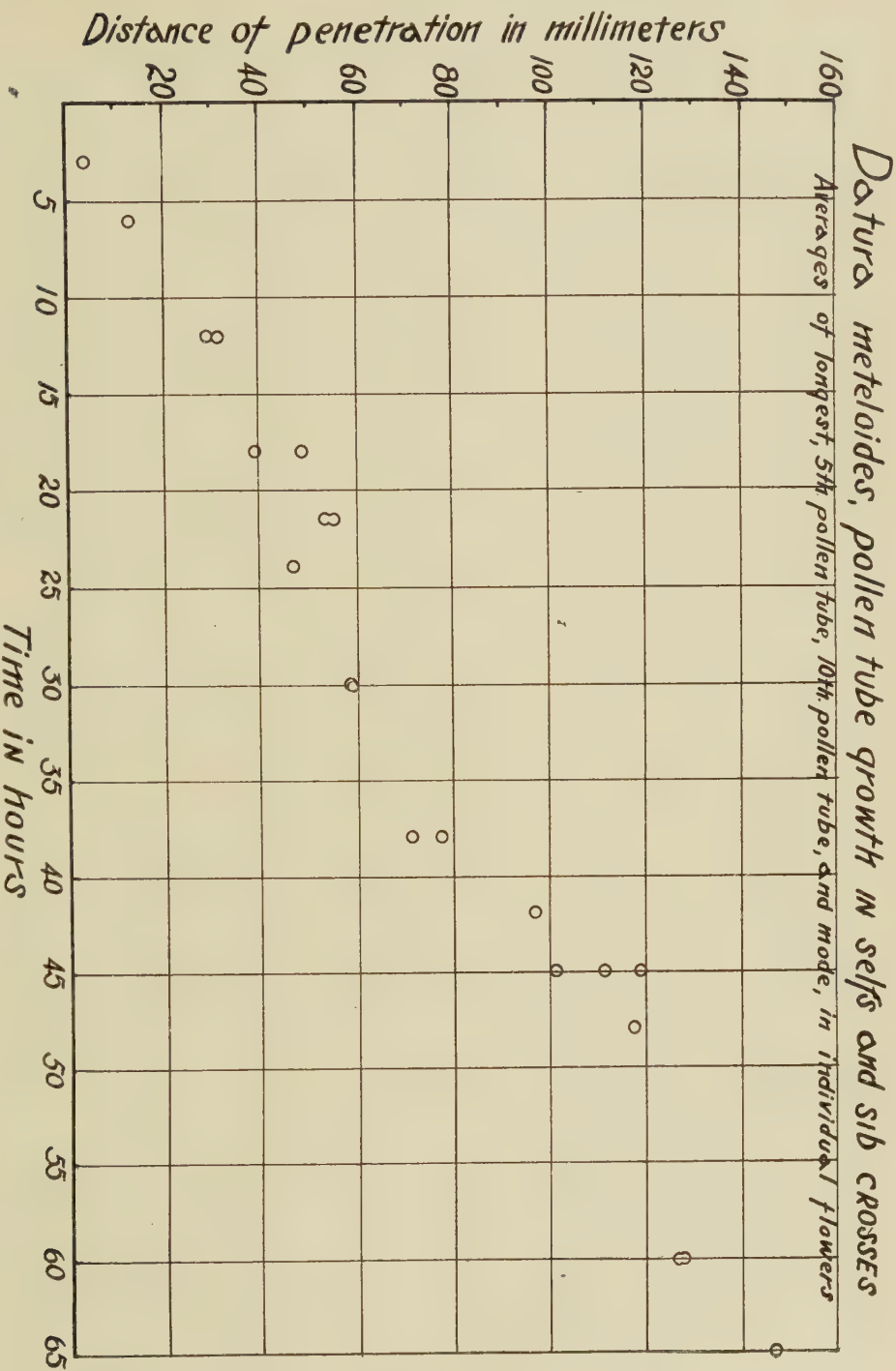


FIG. 3. Graph for pollen-tube growth in *Datura Meteloides* at 17.8° C.

## TEMPERATURE AND POLLEN-TUBE GROWTH

We have also made several sets of determinations on the effect of temperature on pollen-tube growth by the use of this technique. The more complete data obtained from these investigations on temperature are being published elsewhere (2), and we wish here only to present a summary of the results. The distance of penetration of the foremost group of pollen tubes into the stylar tissue was measured with the result that at 11.1° C. the pollen tubes grew at the rate of 1.28 mm. per hour, at 17.8° C. the rate was 2.65 mm. per hour, at 25° C. it was 4.59 mm. per hour, at 28.9° C. it was 5.25 mm. per hour. Near 33.3° C. was the optimum temperature at which the rate of growth was 5.86 mm. per hour with a slight lowering of the rate at 37° C. to 5.72 mm. per hour. Thus we can say that temperature has a very pronounced effect on the rate of pollen tube growth. At the temperature which was nearest the optimum (33.3° C.), the growth rate was four and one-half times as great as at the lowest temperature (11.1° C.), which was tested. At the lowest temperature, a style 60 mm. long could still be traversed completely in less than 48 hours, which is long before abscission occurs, especially when we consider the fact that cool weather usually prolongs the period of duration of the style.

## INTERSPECIFIC CROSSES

One of the interspecific crosses which we have frequently attempted to make, but without success, is the one between *Datura Stramonium* and *D. meteloides*. It was our purpose in making this cross to discover how the incompatibility which exists between these two species manifests itself in relation to pollen-tube growth. These experiments were also carried on at a temperature of 17.8° C.

The cross *Datura Stramonium*  $\times$  *D. meteloides* results in a remarkable uniformity of pollen-tube growth. Just as in the former species where self-pollinated, so in this cross, using the pollen of *D. meteloides* on the stigma of *D. Stramonium*, we obtained almost perfect germination of the pollen. The measurements made on this material are presented in TABLE 2 and the graph showing the effect of time on pollen-tube growth is shown in a graph in FIG. 4. It will be seen that the time required for germination is greater in this cross and that the growth rate is slightly less, but the time is still much greater than is necessary for fertilization.

At 17.8° C. fertilization would take place in from 29 to 33 hours, depending upon the length of the style. The growth rate (1.9 mm. per hour) is as great at this temperature as it would be for sib crosses or selfs of *D. Stramonium* taken at a slightly lower temperature (for example the latter taken at about 14.5° C.) and the sterility of this cross, made with *D. Stramonium* as the female parent, is clearly due to causes other than the pollen tube growth.

We have some other facts gained from a previous attempt to make this cross. From several greenhouse plants pollinated in this way we obtained some seed-like structures.

TABLE 2  
 POLLEN-TUBE GROWTH IN THE CROSS *Datura Stramonium* × *Datura meteloides*<sup>\*</sup>  
 DURING VARIOUS TIME INTERVALS

TIME INTERVAL	DISTANCE REACHED BY LONGEST POLLEN TUBE, MM.	DISTANCE REACHED BY 10TH POLLEN TUBE, MM.	DISTANCE REACHED BY 20TH POLLEN TUBE, MM.	DISTANCE OF APPROX- IMATE MODE MM.	AVERAGE OF ALL MEASURE- MENTS	CORRECTED AVERAGE (x 1.06)
2 Hours						
25P130.....	1.3 1.3	1.1 1.0	1.1	1.0		
Average.....	1.3	1.1	1.1	1.0	1.1	1.2
4 Hours						
25P131.....	4.6 4.4	4.3 4.2	4.2 4.1	4.0 3.8		
Average.....	4.5	4.3	4.2	3.9	4.2	4.5
6 Hours						
25P74A.....	8.1 8.5 8.3	7.6 8.2 8.2	7.4 7.9 7.9	6.7 7.4 7.2		
Average.....	8.3	8.0	7.7	7.1	7.8	8.3
12 Hours						
25P75.....	20.0 19.2 20.4 20.1	19.4 18.2 19.5 19.6	19.1 17.9 19.0 19.3	18.2 15.6 18.2 18.6		
Average.....	19.7	19.2	18.8	17.6	18.9	20.0
18 Hours						
25P74.....	35.3 33.8 33.8	33.7 31.2 33.0	32.5 31.5 32.5	31.4 30.4 29.2		
Average.....	34.3	32.6	32.2	30.3	32.3	34.2
24 Hours						
25P76.....	45.6 44.7	44.0 42.6	42.6 41.9	36.0 39.1		
Average.....	45.1	43.3	42.3	37.6	42.1	44.6
27 Hours						
25P77.....	51.1 48.8 51.7	49.1 47.3 49.3	47.6 46.4 47.9	46.7 44.7 46.9		
Average.....	50.5	48.6	47.3	46.1	48.1	51.0

FIG. 5 shows some of these seeds at the left beside some normal seeds of *D. Stramonium* shown at the right. Among the seeds obtained from the cross were two which were fully enlarged to the size of those of *D. Stramonium* (one of them is shown in the photograph) but these large seeds, as well as the small ones, were all light in weight and empty. None of these seeds germinated.

Evidently the pollen tubes entered the micropyles of the ovules and supplied the stimulus necessary for ovule development. Perhaps the endosperm started to form, which might account for the enlargement of the ovules, but without viable embryos the seeds were left unfilled.

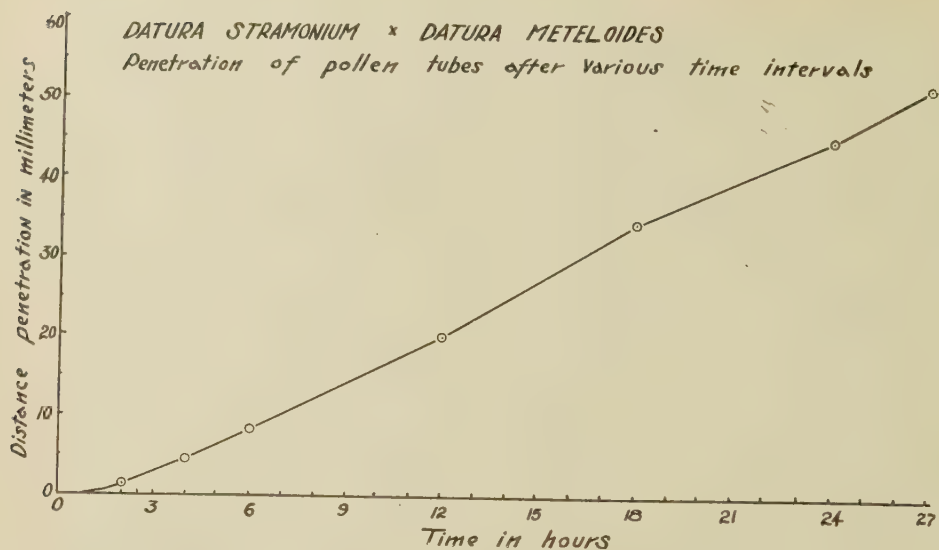


FIG. 4. Graph showing penetration of pollen tubes when *Datura Stramonium* is pollinated with pollen of *Datura meteloides* at 17.8°C.

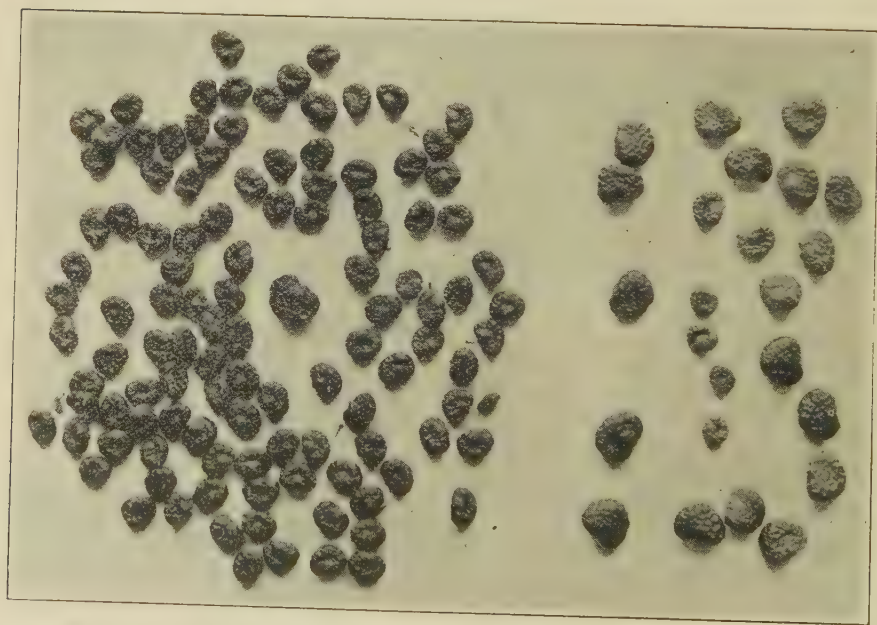


FIG. 5. Abortive seeds at left compared to viable seeds at right. The former were obtained by crossing *Datura Stramonium* with pollen from *Datura meteloides*. Seeds at right are normal seeds of *Datura Stramonium* (including a few abortive seeds of this species).

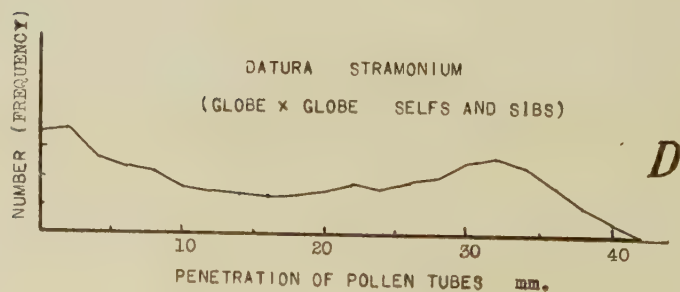
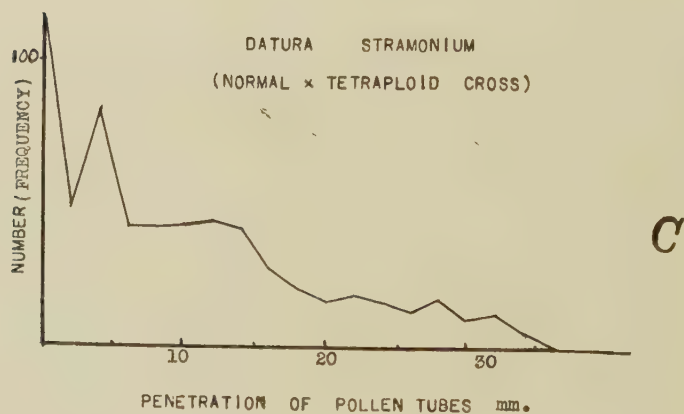
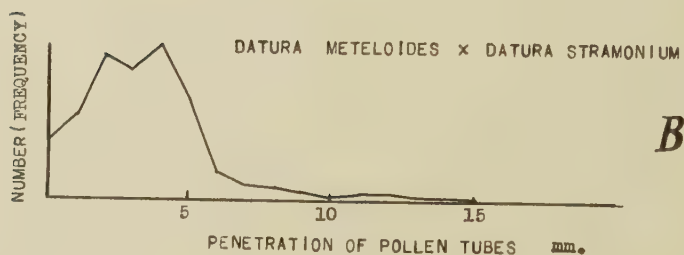
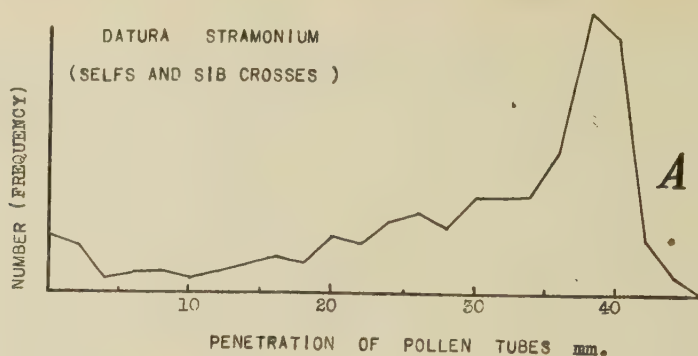


In the reciprocal cross, *Datura meteloides*  $\times$  *D. Stramonium*, the situation is entirely different. Here poor pollen-tube growth is the primary factor which prevents fertilization. The pollen tubes grew so slowly and irregularly that we were not successful in obtaining from measurements data which were worth tabulating. This experiment was also repeated without success on plants in the garden. For example, in the garden the pollen tubes were found to have penetrated only 10 mm. in 24 hours, and at the temperatures used in our other experiments (17.8° C.) the growth was proportionately less. With 160-170 mm. as the distance to be traversed in order to reach the ovary, it is obvious that these pollen tubes could never accomplish fertilization during the life of the flower.

In the latter instance of the cross between these species using *D. meteloides* as the female parent we observed another phenomenon which has not been previously described as far as we are aware, that of a distinct swelling of the pollen tubes. In *Datura Stramonium*, selfed or crossed by sibs, usually only a few pollen tubes in the region near the stigma are swollen, but in this cross in which the pollen tubes are responsible for the incompatibility we find all or nearly all of the pollen tubes abnormally enlarged or swollen (see PLATE 18). Their tips are frequently imbedded in or surrounded by a group of deeply stained cells derived from the conducting tissue, which renders their position much more conspicuous. This condition, namely, the swelling of the pollen tubes may be taken as an index of this type of incompatibility in which pollen tubes are responsible.

We have also observed another condition accompanying this incompatibility due to the pollen tubes. This has to do with the curve of distribution of the pollen tubes along the length of the style at some time after pollination. In normal cases as when selfs or sib crosses of *Datura Stramonium* are made, the pollen tubes are distributed in the manner shown in FIG. 6A. (It should be emphasized that in order to obtain representative slides showing these curves it is necessary to apply a moderate amount of the pollen all at one time. Several successive applications of pollen as might happen in the field under conditions of natural pollinations would not always result in a typical curve of this kind. Also, the stigma must be mature and fully receptive. If not, the pollen grains germinate successively rather than simultaneously. Intense crowding also affects the character of this curve.) Here the mode of distribution is near the advancing front of the group of pollen tubes. One may call this a curve with a forward skew. However, when incompatibility is due to unsatisfactory pollen-tube growth the skew is backward. FIG. 6B shows the condition of pollen-tube distribution in the cross *Datura meteloides*  $\times$  *D. Stramonium*. A small number of pollen tubes have grown out far in advance of the mode and this results in a curve with a marked backward skew.

Similar curves with backward skew may be observed in the case of *D. Stramonium* pollinated with pollen from its tetraploid mutant (FIG. 6C). This cross has not been found to set seed, though the reciprocal cross has



been successful and produced a few triploid offspring. This cross, using tetraploid pollen, also gives a very large proportion of the swollen pollen tubes imbedded among deeply staining cells of conductive tissue.

FIG. 6D shows the pollen-tube curve of distribution obtained from the pollen of a Globe, a  $(2n + 1)$  chromosomal mutant. Here the pollen consists of two kinds of grains in approximately equal proportions: those with  $n$  chromosomes, and those with  $(n + 1)$  chromosomes (1). When this pollen is applied to the stigma the pollen tubes coming from grains with  $n$  chromosomes germinate and grow, forming a distribution curve of the type shown in FIG. 6A. The  $(n + 1)$  pollen grains also germinate but grow more slowly, tending to produce a curve of the character shown in FIG. 6B. It will be seen that curve 6A and curve 6B combined will produce a bimodal curve such as we have in FIG. 6D. The latter curve was obtained by combining the pollen tubes from a dozen or more styles grown for an equal period.

In the Globe  $(2n + 1)$  mutant the swollen pollen tubes may also be observed, but the pollen tubes with  $(n + 1)$  chromosomes do not all show this enlarged character. The proportion of swollen pollen tubes is greatest near the stigma and diminishes toward the ovary.

#### Description for FIG. 6 on page 258

Curves of pollen-tube distribution in the styles of *Datura*, after selected intervals. The pollen tubes were growing from left to right. Numbers of pollen tubes observed in each space were plotted as ordinates; distances of penetration from the stigma were plotted as abscissae. The number of ungerminated pollen grains counted on the stigma were plotted on the O ordinate at the left.

A represents a sib cross of *Datura Stramonium* ( $2n$  plants). This is a typical distribution curve in a plant with great self-fertility representing such a pistil as the one shown in the photomicrograph of PLATE 17. This distribution curve has a forward skew.

B represents an incompatible cross: *Datura meteloides* pollinated with pollen from *Datura Stramonium*. This distribution curve has a backward skew.

C represents a cross of *D. Stramonium* with pollen from a tetraploid, a  $4n$  chromosomal mutant. The cross made in this way has also been found to be incompatible, and the curve shown here has a backward skew.

D represents the pollen of a Globe, a  $(2n + 1)$  mutant in selfs and sib crosses, and combines the populations from a dozen styles, which smoothes out the curve. Here the pollen is composed of grains, half with  $n$  chromosomes, and half with  $(n + 1)$  chromosomes. This pollen therefore results in a bi-modal curve. The forward mode is made up very largely of pollen tubes with  $n$  chromosomes, while the second mode is made up very largely of pollen tubes with  $(n + 1)$  chromosomes. Only a small fraction of the  $(n + 1)$  pollen tubes reach the ovary, a fact which accounts for the transmission of the extra chromosome through the pollen to the extent of only about 1% in ordinary crosses: Normal plants  $\times$  Globe plants.

An examination of the tabulation given by East and Park (3) for several incompatible groups of pollen tubes in *Nicotiana* also shows this backward skew in the curve of pollen-tube distribution. Here, then, is a feature of agreement of our results with those of East and Park in their studies of sterility in *Nicotiana*. Perhaps this feature together with the conditions of swollen pollen tubes will serve as a more universal index to the incompatibility than the presence or absence of acceleration in the growth which East and Park reported in *Nicotiana* and which we do not find in *Datura*. However, it must be borne in mind that just as there are forms of interspecific sterility not due to abnormalities in pollen-tube growth, so we

may expect that the behavior of the pollen tubes may not always indicate the existence of sterility. The abnormal conditions which we observed in the distribution and in the appearance of the pollen tubes were found only when the cause of the sterility is to be found in pollen-tube growth itself.

#### ADDENDUM

As we correct the proof on the above paper which was prepared over a year ago and submitted last summer we may add that, on the basis of some more recent investigations, we have reached some rather definite conclusions concerning the nature of the "swollen" and abnormal pollen tubes mentioned here. They appear very similar to those of the tricarpel which we are describing in a concurrent publication (Proc. Nat. Acad. 1927). In the case of the tricarpel, a gene mutant of *Datura Stramonium*, these abnormal pollen tubes may be found in all stages of swelling and bursting. They represent very largely pollen tubes which have ruptured in the region near their ends, and before actual bursting they may appear with enlarged swollen ends. We now feel confident that this diagnosis fits practically all cases of the abnormal and "swollen" pollen tubes described in this paper, and are making this appended statement after a careful examination of these slides. In normal pollinations of either of these species only a small proportion of the pollen tubes burst within the conducting tissue of the pistil, mostly in the region near the stigma, but these are conspicuous and may be seen in PLATE 16. In the interspecific crosses, represented by PLATE 18, nearly all of the pollen tubes have burst and they represent the backward skews of the distribution curves of Figs. 6 B, C and D.

#### LITERATURE CITED

1. Buchholz, J. T., and Blakeslee, A. F., 1922. Studies of the pollen tubes and abortive ovules of the Globe mutant of *Datura*. Sci. N. S. 55: 597-599.
2. Buchholz, J. T., and Blakeslee, A. F. Pollen-tube growth at various temperatures. (Concurrent publication.) Am. Jour. Bot.
3. East, E. M., and Park, J. B., 1918. Studies on self-sterility. II. Pollen-tube growth, Genetics 3: 353-366.

#### EXPLANATION OF PLATES

##### PLATE 16

Photomicrograph of the apical portion (section *A* of insert and about  $\frac{1}{2}$  of the entire style) of a pistil of *Datura Stramonium* from a preparation obtained by the dissection method of a pistil at 15 hours after proper (sib-pollination). This shows the enlarged region of the stigma above continuous with the strand of conducting tissue containing the pollen tubes. The enlargement is about 16 times.

In the insert is shown a pistil of *Datura Stramonium*, natural size, with the segments, *A*, *B* and *C* indicated which are enlarged in the Plates 16 and 17.

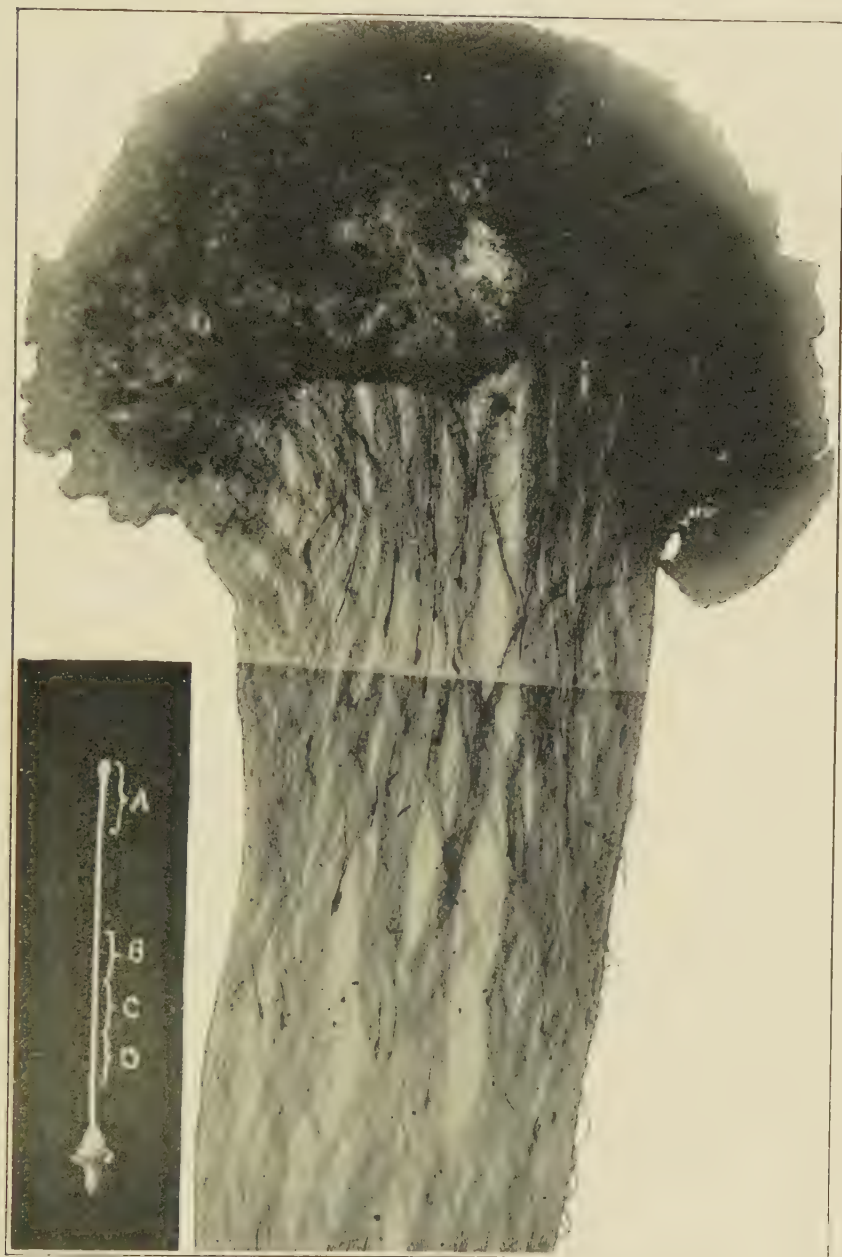
##### PLATE 17

From the same pistil as segment *A* of Plate 16 but from lower down in the strand of conducting tissue in the regions indicated in *B*, *C* and *D* of Plate 16. In *B* the ends of a few scattering pollen tubes will be observed. In *C* the ends of many tubes will be seen at nearly the same level (the mode) with the end of the longest pollen tube somewhat in advance. In *D* immediately below *C* no pollen tubes are to be found. The sections here shown are enlarged 21 times.

##### PLATE 18

Photomicrograph of a dissection of the stigma region of a style of *Datura meteloides* made 24 hours after a garden pollination with pollen of *Datura Stramonium*. The pollen tubes have penetrated to only a short distance and they are swollen and enlarged and are surrounded by deeply stained cells of the conducting tissue. Magnified about 16 times.





BUCHHOLZ AND BLAKESLEE. POLLEN-TUBE BEHAVIOR





BUCHHOLZ AND BLAKESLEE: POLLEN-TUBE BEHAVIOR







BUCHHOLZ AND BLAKESLEE: POLLEN-TUBE BEHAVIOR



# THE ROLE OF STERILITY IN THE IMPROVEMENT OF VEGETABLES

C. E. MYERS

*Pennsylvania State College*

(WITH PLATE 19)

To those familiar with the general principles of genetics little argument is necessary to show the need of self-fertilizing the plants with which they are working. This is of primary importance if one is to make definite progress. For those species which are known as self-fertilized little if any difficulty is experienced, but for those which are normally cross-fertilized, quite a difficult problem confronts us.

The importance of self-fertilizing plants of this type, however, has been clearly and positively shown by Jones (14) in his work with corn. Attempts to improve this crop along definite lines, to conform to a particular standard, date from an exposition held in Chicago in 1886. During the forty years which have intervened various methods of selection have been practiced and specimens approximating the ideal have been obtained. However, none of the various methods practiced have led to the production of the show type ears with any great degree of certainty. Furthermore, many of the prize-winning ears have given mediocre progeny from the standpoint of both appearance and yield. At a symposium on "Present Status of Corn Improvement" during the past year, the Chairman, Dr. F. D. Ritchey (18), called attention to the fact that the evidence from the varietal trials shows few, if any, varieties or strains developed by ear-to-row breeding which are on a materially higher plane of productiveness than the best of the mass selected varieties of the locality. He states, "The consideration of mass selection, ear-to-row breeding, and the desirability of  $F_1$  varietal hybrids, however, was more appropriate to the symposium on corn breeding held by this society five years ago than it is here. The subject today is "The Present Status of Corn Improvement" This must needs exclude from consideration any method not involving some degree of selection within self-fertilized lines. True mass selection or ear-to-row breeding still may be utilized to raise the productiveness of inferior or unadapted sorts to a level approximating that of the better varieties. Mass selection also is an important means of maintaining productiveness in adapted strains. Improvement, however, implies more than this, and it is by methods involving selection within self-fertilized lines that the corn breeder of today is attempting to obtain yields sufficiently larger than those now produced by our best varieties."

It is assumed that the details of the method developed by various investigators, including East and Hayes (7) and Shull (22), and elaborated and perfected by Jones (14) are sufficiently familiar to all. The important fact

to note is that although at first this method was not generally accepted, it is now being considered as the only known method which gives evidence of reaching the desired end. This is because of the fact that it is scientifically correct.

I believe that we, who are concerned with the improvement of those vegetable crops which are normally cross-fertilized, have a problem which is practically identical with that of corn. Obviously, we are dealing with material which is extremely heterozygous, and until some method is devised by which it may be brought to a homozygous condition, little definite progress can be expected.

Before discussing the work with those crops, we may well pause to consider the way the subject of sterility has been attacked and the results which have been obtained by those investigators who were concerned primarily with the physiological aspects of the subject.

In practically every instance which has come to my attention, the crop concerned has proved self-sterile to a greater or less extent. By this is meant that the pollen of the flower is incapable of fertilizing the ovules of that flower or of another flower of the same plant, but that those ovules are capable of setting seed when pollinated with pollen from another plant. Similarly, the pollen functions when used to pollinate the ovules of another plant. Colter (4) states, "In many instances the problem is tied up with that of the growth of the pollen tube. Own pollen, quite healthy and functional on foreign stigmas, will also germinate and start pollen tubes on own stigmas. Such tubes, however, are not successful in reaching the ovules. Assumptions were made that own stigmas poison own pollen tubes, or furnish them with no adequate nutrition." On the other hand, Moore (16), working with three species of *Tradescantia*, including twelve plants, made eighty-three self-pollinations and twenty-six cross-pollinations. None of those selfed produced seed, while all of the crossed did. Cytological studies were made and he concluded that "own stigmas provided own pollen tubes with too good nutrition so that the tube flattened, but did not elongate." He contrasted this with the phenomenon observed in a fungous in which the hypha tends to elongate when grown in a poor medium.

East has done much valuable work on this subject with *Nicotiana*. In conjunction with Park (9), they found that pollen germinated on own stigmas just as well as does foreign pollen, and the first increment of growth takes place at the same time. From that time on the pollen tube tends to grow steadily, but the rate of growth of the foreign pollen tube is considerably accelerated, as though it were receiving some stimulus which is ineffective on own pollen tube. The result is that the own pollen tube fails to reach the ovary before the stigma and style have decayed, while the foreign pollen tube, due to accelerated growth, reaches the ovules. They observed, however, that pollination at the end of the season brought about the desired end, and by this means self-sterile individuals were inbred. East (10) also obtained data from which he concluded that self-sterility in the plant is due to the



presence of a single Mendelian recessive factor. The same worker (12) states, "Sometimes it is possible to so control the conditions that one may obtain seed from a particular mating when it is impossible to obtain it after a mating made the reverse way. For example, if plant A, a plant in vigorous condition and at the height of its flowering season is used as a parent on an incompatible plant B, a plant in weakened condition at the extreme end of the flowering season, some seed will be obtained as a manifestation of pseudo fertility, but a reciprocal cross is impossible."

Stout (23) working with several different species, has made important contributions to the physiology of the subject. In his work with *Lythrum salicaria*, he observed a very decided case of self-compatibility at the close of the period of bloom. In his work with *Brassica pekinensis* and *B. chinensis*, in which more than 1,300 plants were tested, 653 were found to be completely self-incompatible, while 718 plants were self-compatible to some degree. Self-compatibility was strongest during the period of mid-bloom. He finds "that self-compatibility in these species is limited to a specific period following the transition from vegetative to reproductive activity, and limited by waning senility of the plant as a whole. Self-compatibility appears coincidentally with the climax of reproductive activity."

The same writer (24) observes that in some species there is self-compatibility at the end of the period of bloom, in others at the mid-period and, for certain perennials, there is some evidence of change from year to year in relation to the age of the plant. In *Brassica pekinensis* he observed that self-incompatibility may be somewhat influenced by cultural treatment which reduced vegetative vigor. In a family of this species, grown for three generations, less than 10 per cent of the total of 326 plants were highly self-compatible, and there was no hereditary effect of selection for self-compatibility. When a generation of this family was grown in small pots, with a decided reduction of vegetative vigor, of a total of 1,128 plants there were 734, or 65 per cent, that were highly self-compatible and only 22, less than 2 per cent, were self-incompatible. Furthermore, a large percentage of the former were self-compatible in the earliest flowers that opened. The family showed decided change in the number of plants that were self-compatible, and in the individuals the characteristic cycle was altered. The results indicate that there is a direct and very decided physiological correlation between vegetative vigor and the functional properties of the organs concerned with fertilization.

Kakizaki (15) worked with Pe-tsai, which normally produces seed from almost every flower. In his experiment he found that flowers pollinated with their own pollen, those pollinated with that from different flowers in the same inflorescence, and those pollinated with pollen from different inflorescences of the same plant gave approximately similar results. He observed that about 30 per cent of the flowers treated produced normal pods, 20 per cent produced imperfect pods, while the remainder were entirely sterile and failed

to develop, although flowers pollinated by other plants gave approximately 96 per cent of fertility.

With respect to work along this line with economic plants aside from corn, that with cucurbits has probably proved most fruitful.

The literature with respect to self-fertilization of this crop is conflicting. Bailey (1), in 1900, reported on a large number of attempts to self-fertilize squash and pumpkins, but failed, and concluded that his material must be self-sterile. In 1904 Cummings (5) reported that he successfully selfed two varieties of *Cucurbita pepo*.

Later Cummings and Stone (6), working with the Hubbard squash, found that three consecutive years of self-fertilization did not tend to lessen the yield. By this method, both high and low yield strains were obtained. The high producing strain produced 12 per cent more squashes, weighing 44 per cent more, the average squash being one-fourth heavier.

Bushnell (2) has also worked with the Hubbard squash. It is generally heterozygous for fruit characters. Because of this lack of uniformity, an attempt was made to self-pollinate a number of plants for several generations, as a means of securing uniform strains. In 1914 an attempt was made to self-fertilize a number of plants, but only two fruits matured. The work was repeated in 1915 and forty-seven fruits were matured. The seeds from these were planted in 1916 and fifty-eight matured fruits were obtained; similarly in the following year, 117 selfed fruits were obtained. Thus far, however, since selfed fruits had not been obtained from other plants; it was possible that in such heterozygous material some plants were sterile. In 1918 only two plants of each strain were used and an attempt was made to self every pistillate flower appearing in the plot. In this way selfed fruits were secured from every plant. Similar results were obtained in subsequent years. Bushnell observed that successful pollinations could be made under a considerable range of conditions with respect to weather; likewise successful pollinations were made at every hour of the day, from 6 A. M. to 6 P. M. The highest percentage of successful pollinations were made when the petals were widely spread.

A study of the flowering habit and sequence of flower production of ten plants which he studied indicated that at certain periods the flowers set if pollinated, while at other times during the blooming period all the flowers abort. This sterility is most pronounced during the early part of the flowering period. In a subsequent paper (3) he reports on the gain made in producing a more uniform type of fruit by inbreeding. As a result of this work, in 1921 a small-fruited strain which produced fruits averaging 2.4 pounds was placed on the market. It has been named the Kitchenette Hubbard.

Rosa (19), working with the cantaloupe, observed the reverse condition to be true with respect to the time of compatibility during the blooming period. He found that practically none of the numerous perfect flowers could be made to set fruit during the middle portion of the growing season, regardless of the method of pollination. Usually two perfect flowers are produced

on a spur. A study of these showed that 42 per cent of the first spurs on each branch set and develop fruit, while only half as many set and develop in the second and third periods. He found that in this crop there are two and sometimes three periods when fruit setting takes place. The same investigator (20) worked with watermelons. He bagged eighty-one hermaphrodite blossoms, but not one matured fruit. He selfed other flowers and the result was that fruit matured, with a normal amount of seed.

At the Horticultural Experiment Station, at Vineland, Ontario (17), work has been in progress for several years, the aim being to develop a strain of cucumber which need not be pollinated by artificial means. The original parents were White Spine and the English Long type. The latter was used because it fruits quite freely under greenhouse conditions, and without artificial pollination. Subsequent selection has been practiced for ten generations for free setting, with the result that "several desirable types of greenhouse cucumbers have been obtained, types which are apparently capable of setting fruit very freely without fertilization."

Shaw (21) made some studies on self-, close- and cross-fertilization in beets. Some workers have held that this species is self-fertile. When Shaw inclosed single plants under closely woven cloth, having sixty-four threads to the inch, such as had been used by the European investigators, he obtained about 23 per cent of fertilization. Separate flowering stems isolated with paper bags generally remained completely sterile. He concluded that the material used in the cloth bags permitted a considerable amount of foreign pollen to be admitted, which produced the seeds that were obtained. He did, however, obtain from 5 to 8 per cent of fertilization when the pollen of the flowers of one spike was placed on the stigmas of flowers of another spike on the same plant, while under similar conditions self-pollinated pistils proved sterile. Hammerlund (13) in his investigations has found that self-sterility is very unequal for different strains of cabbage, and seems also to vary for other kinds of green vegetables.

For some years I have been interested in the improvement of cabbage. This has necessitated attempts to self-fertilize those plants with which I have been working. In practically every case, however, my efforts have been fruitless. Work has been conducted both out of doors during the month of June and in the greenhouse during the month of April. As typical of the work of other years that for 1924 may be cited. That year the field work was centered on twenty-seven plants, each of which was covered with a cheesecloth bag. Close pollination was practiced; that is, all the blossoms of the plant which were shedding pollen were worked with a camel's hair brush almost daily during the blooming period, which lasted about three weeks. Each time the apparatus was sterilized with alcohol before proceeding to the next plant. In all, 3,732 blossoms were worked, but practically no seed was obtained. However, individual blossoms crossed by pollen from sister plants produced an abundance of seed from both direct and reciprocal crosses.



During the past winter, eight plants of the same variety were grown in the greenhouse. When they came to the flowering period, various attempts were made to self-fertilize them under different conditions, such as time of day, amount of sunlight, stage of blossom development, and under various conditions with respect to moisture. In some instances the plant was subjected to a lack of water and was decidedly wilted, while in other cases the plant was given an abundance of water. In every instance ample pollen was present, but in no case was seed obtained from the selfed blossoms. Reciprocal crosses with sister plants, on the other hand, gave a splendid set of seed.

However, the results of not all of those who have worked on the sterility of this crop have been as unproductive as have mine. Dr. C. H. Myers, of Cornell University, Ithaca, has informed me that he has had quite satisfactory results from selfing plants under both field and greenhouse conditions; likewise Professor L. R. Detjen, of the University of Delaware, has been similarly successful. I am at a loss to know why they have succeeded and I have failed, but we have not abandoned the search for the reason, and ultimately hope to find the solution to the problem.

#### BIBLIOGRAPHY

1. Bailey, L. H., 1890. Experiments in crossing cucurbits. Cornell Agric. Exp. Sta. Bull. 25.
2. Bushnell, J. W., The fertility and fruiting habit in Cucurbita. Soc. Hort. Sci. Rept. 17: 47-51.
3. ——— 1923. Isolation of uniform types of Hubbard squash by inbreeding. Soc. Hort. Sci. Rept. 1922: 139-144.
4. Coulter, M. C., 1923. Outlines of genetics. University of Chicago Press.
5. Cummings, M. B., 1904. Fertilization Problems. A study of reciprocal crosses. Maine Agr. Exp. Sta. Bull. 104. 81-99.
6. Cummings, M. B., and Stone, W. C., 1921. Yield and quality in Hubbard squash. Vt. Agr. Exp. Sta. Bull. 222.
7. East, E. M., and Hayes, H. K., 1911. Inheritance in maize. Conn. (New Haven) Agr. Exp. Sta. Bull. 167.
8. East, E. M., and Park, J. B., 1917. Studies in self-sterility. I. The behavior of self-sterile plants. Genetics 2: 505-609.
9. ——— 1918. Studies in self-sterility. II. Pollen tube growth. Genetics 3: 353-366.
10. East, E. M., 1918. Studies in self-sterility. III. The relation between self-fertile and self-sterile plants. Genetics 3: 341-345.
11. ——— 1919. Studies in self-sterility. A family of self-sterile plants wholly cross-sterile inter-se. Genetics 4: 356-363.
12. ——— 1923. Genetical aspects of self- and cross-sterility. Amer. Jour. of Bot. 10: 468-473.
13. Hammerlund, H. C., 1920. Improvement of green vegetables at the station for plant improvement of Weibullsholm Landskrona (translated title). Bot. Abstr. 5: Entry 387.
14. Jones, D. F., 1920. Selection in self-fertilized lines as the basis for corn improvement. Jour. Amer. Soc. Agron. 1920: 77-100.
15. Kakizaki, Y., 1922. Self-sterility in Chinese cabbage. Jour. Heredity 13: 374-376.
16. Moore, C. W., 1917. Self-sterility. Jour. Heredity 8: 203-207.
17. Ontario Dept. of Agr. Rept. of Hort. Exp. Sta., Vineland Sta. 1919: 18-20.
18. Ritchey, Frederick D., 1926. Jour. Amer. Soc. of Agron. 18: 306.
19. Rosa, T. J., 1925. Pollination and fruiting habit in the cantaloupe. Proc. Soc. Hort. Sci. 1924: 51-57.
20. ——— 1926. Pollination and fruiting habit in the watermelon. Rept. Soc. Hort. Sci. 1925: 331-333.
21. Shaw, H. B., 1916. Self-, close- and cross-pollination of beets. Mem. N. Y. Bot. Gard. 6: 149-152.
22. Shull, G. H., 1911. The genotypes of maize. Amer. Nat. 45: 234-252.
23. Stout, A. B., 1922. Cyclic manifestation of sterility in *Brassica pekinensis* and *B. chinensis*. Bot. Gaz. 73: 110-132.
24. ——— 1923. The physiology of incompatibilities. Amer. Jour. Bot. 10: 459-61.





A self-incompatible plant of Danish Ballhead cabbage. Capsules formed to self-pollination but these contained no seeds. Flowers cross-pollinated with a sister plant set capsules containing numerous viable seeds as shown at the left.



## NUTRITIONAL FACTORS IN SEED AND FRUIT FORMATION IN VEGETABLE CROP PLANTS

PAUL WORK

*Cornell University*

The term sterility describes the failure of a plant to produce viable seed. Such failure may occur at many different points in the development of either gamete and its associated parts or of the embryo after fertilization. The delicate processes of gamete development, of pollination and of fertilization are subject to a wide variety of surrounding conditions, a large group of which may be embraced under the general concept of nutrition. Here are involved not only the supply of nutrient materials from without, but also the processes of metabolism within the plant which affect the availability of the nutrients for various uses. Fruitfulness and sterility themselves profoundly influence nutritional conditions within the plant.

Investigation of internal conditions associated with external treatments and with external manifestations of plant performance is still in a primitive state although it has received much attention during the past decade. Work that has been carried out with cabbage and celery is reported to this conference in other papers. The present discussion will be pretty largely confined to the tomato. Findings with this plant are highly suggestive in their applications to other crops though generalizations must be attempted with great caution in view of the wide variations in the nature of plants as, for example, lettuce, beets, peas and melons.

Sterility in the tomato is a problem of prime practical significance. In hot climates, heat and drouth seem to interfere with fruit set as also does irrigation at the wrong stage of plant development. Rainy weather at setting time has the same effect. Insects, such as thrips, are accused of interfering with pollination. Actual shortage of nutrient elements in the field reduces the set of fruit. Certain varieties are more subject to the conditions occasioning "running to vine" whether this is the cause of failure to set fruit or whether it is a result.

The history of gamete development and of pollination and fertilization have not been thoroughly studied cytologically in the tomato, but gross observation shows that floral primordia may remain small and drop off or flowers may develop normally to all external appearance and yet fail to set fruit. We need fuller knowledge of the natural history of the tomato plant.

The classic researches and speculations of Klebs (1, 2) may be regarded as having laid the foundation for the present interest in the relation between nutrition and sterility. Practical problems crying for solution have inspired the builders who have followed with detailed studies. Year after year

Klebs experimented with the many devices by means of which he was able to influence the performance of plants as regards vegetation and reproduction. While he was engaged in these studies other experimenters were content to apply many different treatments to the soil and to observe the consequent behavior of the plant, bestowing little consideration upon the chain of internal causes that Klebs emphasized as connecting any primary treatment or condition of environment with its resultant manifestation in plant performance.

The concept of external factors modifying performance through their effect upon internal conditions was an epoch-making contribution to plant research, but Klebs did not offer experimental evidence as to the nature of the internal causes. It remained for Kraus and Kraybill (3) to throw definite light upon the conditions that are intermediate between the external treatments and the external evidences of plant performance. In so doing they inaugurated an era of investigation that has yielded more new knowledge of nutrition than any comparable movement certainly in horticulture and perhaps in all plant science. Among the numerous factors that make up the internal economy of the plant, their work called attention to carbohydrate and nitrogen content as important links in the causal chain, and they pointed to certain relations between these constituents corresponding to certain relations between vegetative growth and reproductive activity. Their readers have gone further and have postulated a CH/N ratio as a factor conditioning plant behavior, though the authors themselves did not do so and have since warned against anything approaching mathematical application of the ratio idea. Such interpretation is illustrated in a review by Knight (4).

"Thus with the magnitude of the carbohydrate-nitrogen (CH/N) ratio is associated a definite type of growth. Low ratio results in vegetative growth and very high ratio reduces vegetative growth without inducing fruitfulness; when the value is intermediate, a big plant is obtained."

Work (5) approached the problem from another angle, his earlier studies being concerned with an effort to learn something of the nitrogen requirement of the tomato. This led to quantitative nitrate treatments in quartz sand, to careful quantitative records of performance and to determinations of carbohydrates and nitrogen in the plants. Applications of nitrogen, almost heavy enough to plasmolyze the plant, did not induce a condition of high vegetation at the expense of fruitfulness though other varieties than the one used (Bonny Best) might have so reacted. Nitrate of soda did not manifest toxic effects in heavy applications but injury seemed to be due to effect upon availability of soil moisture setting up a condition of "physiological drouth."

Analyses of plant material suggested intimate relation between nitrogen content of the plant and its performance as regards both vegetation and fruitfulness. There was no indication that either high or low carbohydrate content inhibited either vegetative growth or fruit making, nor did a comparison of nitrogen and carbohydrate content in the leaves (green weight basis) indi-



cate any correlation between the two. The nutritional relations seemed better explained on the theory that nitrogen content was the limiting factor and that the carbohydrate content was the resultant of the various processes of utilization against the process of manufacture. This does not minimize the importance of carbohydrates, but merely points to the fact that, barring culture in reduced light or with greatly reduced leaf surface, the available carbohydrate supply did not fall below current needs. Carbohydrate manufacture begins at once on germination and is one of the first activities to revive after a period of nitrogen starvation. When low nitrogen limits growth, photosynthesis does not cease at once and unused carbohydrate material goes into storage, giving rise to high figures. Plants with carbohydrate contents ranging from .92 per cent to 3.66 per cent were in a state of vigorous growth and reproduction.

The concept of the limiting factor has proved highly useful in the interpretation of field experimental results. In fact, the general practice in experimentation of varying one factor at a time and providing a favorable excess of all others rests upon this concept.

While most writers have avoided taking the ratio idea too literally, the question arises whether this concept has not been overworked. The whole implication is that growth at a given moment depends upon a combination of two factors.

A ratio between two factors may be varied greatly without affecting the response of the plant. "Let us assume a plant so situated that all factors for its activity are in favorable excess except nitrogen and carbohydrates. Let nitrogen be available in an amount sufficient to permit half the growth of which the plant is capable. With carbohydrates absent, there will be no response. Now let carbohydrates become available in successive increments. We may expect the plant to respond according to these increments, rather than in proportion to the changing ratio of CH/N, until a point is reached such that the nitrogen available can be fully utilized. Further increments of carbohydrates will continue to change the ratio, but will not be of service to the plant and its growth will not respond further. Assuming now that carbohydrates have become available up to the full need of the plant, increments of nitrogen will be effective according to their amount until the plant has attained its maximum possibility. Thus, in general, plant performance is to be related, not to the ratio between two possible limiting factors, but to the available amount of the single factor that is in minimo at that moment." There appears little reason why we should not consider any number of relationships between single factors as governing growth and reproduction or for that matter triplets or quartets of factors. At least we may well content ourselves, as far as possible, with the effects of a single factor at a time until our knowledge of these individual factors and their influences is much fuller than at present.

The figures cited above suggest that a small content of carbohydrate is sufficient for the plant provided the supply is constantly replenished. Before

we can fully understand plant metabolism we will need to concern ourselves with *rates* of manufacture and use as well as with *content*. We need chemical "movies" in addition to the "stills" which our usual analyses afford. Thus there may be a profound change in the relation between carbohydrate and nitrogen content with no change in effect provided the rate of supply is ample. In other words, an increase in rate of photosynthesis may readily outweigh a decrease in relative amount of carbohydrate to nitrogen present at a given moment.

Kraus (6) has said in commenting upon Murneek's work, "Just as soon as nitrogen is restricted, vegetation decreases, carbohydrate accumulates a later fruit setting and even differentiation of flower is stopped." This would seem to afford a clearer picture of the relationship than attributing change in performance to a change in ratio.

Murneek (7), in what seems the outstanding contribution to this field since Kraus and Kraybill, presents a study of the effect of fruition upon vegetation in the tomato. Work (5) observed what he called "waves of fruitfulness" manifested in an absence of fruit on the central clusters of certain plants which were limited in their nitrogen supply. After the fruit of lower clusters matured, upper clusters bloomed and developed normally. This phenomenon Murneek has studied exhaustively and his conclusions point to nitrogen as the usual factor which is somehow or other placed in *minimo* by the presence of developing fruit. There seems to be a definite relation between the nitrogen resources of the plant and the amount of fruit required to effect the limitation of vegetation.

Any essential factor may be the limiter if it is not present in sufficient amount. Evidence available to date points to the outstanding frequency with which nitrogen assumes this limiting role. Nitrogen is present in plants in many forms. In amino acid form it is the characteristic building stone of the proteins which function in many ways. There is evidence that nitrogen is relatively plastic, changing readily from one form to another. Hitherto we see little conclusive evidence as to what form of nitrogen is most often the determining factor in plant reactions. The importance of the amino acids has been emphasized in this connection. Nitrates, as the form in which nitrogen enters the plant, is much noticed in this connection. Investigations along this line are gravely handicapped through the crudeness of our analytical methods. The time will doubtless come when we are brought to a comparative standstill under our prevailing experimental methods and we may be forced back to less spectacular and more laborious successive analyses dealing with many parts of plants, perhaps tracing the amino acid or the nitrate content week by week through a season of growth. The worker who develops rapid and reliable quantitative methods applicable to small samples will be a great benefactor.

Attention may well be called to the necessity of considering internal relations in plants from any angles. The curves for nitrogen and carbohydrate content in Work's (5) study show very clearly the absence of a correlation

between these two factors. The basis is green weight in leaves, chosen because the green condition is the active condition of the plant and because the leaves were deemed to be the principal workshop of metabolism. Roberts (8) bases curves on figures from the same publication, but using instead, dry weights of stems. These curves show on their face a very pronounced negative correlation. Suppose, however, we consider the diagram in two sections, the first representing plants with low nitrogen content resulting from low nitrogen supply; the other, plants with medium nitrogen supply. At the point of division between these two sections there is a very sudden rise in the nitrogen curve. A smoothing of the nitrogen trace in each section would give two lines, each almost horizontal, thus suggesting no correlation at each of the two levels. Even leaving out of account the relative significance of green weight of leaves and dry weight of stems, one cannot but wonder whether the negative correlation interpretation or the no correlation interpretation is more valid. This example illustrates the necessity for the gathering of data in a wide diversity of forms and for its interpretation in many ways. We need to know the total amount of a given material in the plant as well as its concentration, its form as well as its quantity. We must guard against distortions such as occur in green weight figures for moribund plants with their low moisture content. So might examples be cited almost *ad infinitum*, nor do such considerations leave us cock-sure.

Truly our problem is most complex. The weakness of our analytical methods and the narrow limitations of possible experiment, the difficulties involved in precise control of conditions sometimes discourage almost to despair, but scientific experience bolsters the faith that persistent inquiry will gradually write our answers and in this faith we plod on.

#### LITERATURE CITED

1. Klebs, Georg, 1903. Willkürliche Entwicklungsänderungen bei Pflanzen, pp. 1-166.
2. ——— 1910. Alterations in the development and forms of plants as a result of environment. Roy. Soc. London, Proc. **82B**: 547-558.
3. Kraus, E. J., and Kraybill, H. R., 1918. Vegetation and reproduction with special reference to the tomato. Oregon Agr. Coll. Exp. Sta. Bull. 149.
4. Knight, R. C., 1924. The carbohydrate nitrogen ratio. Science Progress **19**: 34-42.
5. Work, Paul, 1924. Nitrate of soda in the nutrition of the tomato. Cornell Univ. Memoir 75.
6. Kraus, E. J., 1925. Soil nutrients in relation to vegetation and reproduction. Amer. Jour. Bot. **12**: 510-516.
7. Murneek, A. E., 1926. Effect of correlation between vegetative and reproductive functions in the tomato. Plant Physiology **1**: 1-56.
8. Roberts, R. H., 1926. Apple physiology. Wis. Res. Bull. 68.





## ENVIRONMENTAL FACTORS AFFECTING SEED-STALK DEVELOPMENT IN CELERY

H. C. THOMPSON

*Cornell University*

(WITH PLATES 20-21)

Celery started in the greenhouse or hotbed, and set in the field as soon as weather conditions permit, frequently goes to seed long before the plants reach full size. Occasionally late celery, from seed sown out of doors, produces seed stalks the same season. This so-called premature seeding results in considerable loss to growers and this fact led the writer to undertake a study of the problem. A summary of the results of the experiments of the first five years has been published (Thompson, 1923) and need not be given in detail here. The factors on which data have been secured include the effects on seed-stalk development of (1) time of sowing seed, (2) checking growth of seedlings by withholding water, by allowing them to become crowded in the flats and by freezing, and (3) by subjecting plants to relatively low temperature for 2 to 4 or 5 weeks.

To determine the effects of time of sowing seed on seed-stalk development, four plantings were made each year: December 10, January 10, February 10 and February 25 or March 10. Plants from all dates of sowing were set in the field at the same time, during the latter part of April or the first few days of May. In every instance the plants from seed sown December 10 and January 10 began to develop seed stalks in June, while those started at the later dates usually did not produce them at all. However, in some seasons a few "seeders" developed in the plants from the two later sowings. The average percentage of seed stalks for the four dates of sowing during four years 1919 to 1922 were 92.10, 75.85, 36 and 3.25 per cent for December 10, January 10 and February 25 or March 10 respectively. Plants from the two early sowings produced ripe seed.

Checking growth by withholding water, by crowding the seedlings in the flats and by freezing materially delayed the development of the seed-stalk every year and with plants from all sowings as is shown in tables 2, 3 and 4 of the report previously mentioned.

The temperature to which the plants are subjected during the early stage of their growth seems to have a very marked effect on seed-stalk development. This was suggested by Whipple (1915-1919) and later by the writer (1923) and by Starring (1924). In 1923 and 1924 the writer subjected some of the plants to relatively low temperature in the cold frame for periods of 17 to 35 days prior to setting them in the field. In 1923 plants from the December and January plantings only were subjected to the low temperature in the cold frame while in 1924 some from all four dates of sowing

were included. In 1923 the plants from the first two were in the cold frame from April 10 to April 27, while in 1924 those from the same dates of sowing were in the frame from April 10 to May 3, and those from the last two sowings were subjected to the cold-frame treatment from April 10 to May 14. Comparable lots of plants were, in all cases, kept in the greenhouse until time for planting in the field as a check against the cold-frame treatment.

The results of these experiments are shown in the following table:

TABLE 1  
EFFECTS OF SUBJECTING CELERY PLANTS TO RELATIVELY LOW TEMPERATURE ON SEED-STALK DEVELOPMENT

DATE OF SEED SOWING	TEMPERA- TURE TREAT- MENT	TOTAL NUMBER PLANTS	NUMBER AND PERCENT OF SEED STALKS IN A GIVEN NUMBER OF DAYS FROM FIELD PLANTING					
			39 DAYS		90 DAYS		120 DAYS	
			NO.	%	NO.	%	NO.	%
12-10.....	Low	212	115	54.24	207	97.64	210	99.05
12-10.....	Medium	249	82	32.93	228	91.56	235	94.37
1-10.....	Low	213	2	0.94	183	85.91	203	95.35
1-10.....	Medium	248	1	0.40	101	40.72	134	54.03
2-10.....	Low	144	0	0.00	56	38.88	70	48.61
2-10.....	Medium	144	0	0.00	0	00.00	0	00.00
2-25.....	Low	144	0	0.00	43	29.86	76	52.77
2-25.....	Medium	144	0	0.00	0	00.00	0	00.00

During the time the plants were in the cold frame in 1923 the mean temperature was about 45° F. and in the greenhouse it was about 60°. In 1924 the mean daily temperature in the cold frame averaged 53.3° from April 10 to May 3 and in the greenhouse 68.3° for the same period. During the period April 10 to May 14 the daily mean in the cold frame was 57° and in the greenhouse 69.4° F. In other words, the temperature averaged 12 to 15° lower in the cold frame than in the greenhouse during the time the plants were subjected to the so-called low-temperature treatment. The temperature never reached freezing in either the cold frame or greenhouse, but at night it went below 40 in the cold frame about half the time.

The significant fact brought out in the table is in connection with the plants from the February 10 and 25 sowings. The plants kept in the greenhouse until set out May 15 did not produce a single seed stalk, while approximately 50 per cent of those which had been subjected to relatively low temperature in the cold frame from April 10 to May 14 not only developed seed stalks but matured seed as shown in PLATE 21.

Additional evidence on the effect of exposure to relatively low temperature on seed-stalk development is furnished by the experience of commercial growers in 1924. Many growers suffered serious financial loss that year due to premature seeding of their celery although they used the same strain of seed and grew their crop in the same way as in previous years. Weather Bureau records at Ithaca show a daily deficiency of 2.6° F. for April and 5.9° for May that year as compared to normal. In one instance, from which

accurate records were secured, a grower sowed seed in a greenhouse and in the open at the same time, about the middle of April, using seed from the same package. The plants from these two sowings were set out in June. Those started in the greenhouse showed no evidence of going to seed at harvest time in September while nearly all of those started outside produced seed stalks three feet high. Many other illustrations might be given.

Studies of the effects of temperature on seed-stalk development also have been made under somewhat controlled conditions in the greenhouse. Plants from seed sown Aug. 28, 1925, were subjected to relatively low temperature in the cold frame for 10, 20 and 30 days from October 16 to November 15. Some plants were also kept at relatively high temperatures (70°F) for 30 days—from October 16 to November 15. Comparable lots of plants were kept at medium temperature as a check. Prior to the temperature treatment all plants had been kept at medium temperature (cool greenhouse). After the treatment the plants were potted and some were placed in a warm greenhouse where the temperature averaged around 70° F. and others in the cool greenhouse where the temperature averaged about 60°.

The temperature averaged 47° F. in the cold frame, 58.8° in the cool greenhouse and 69.4° F. in the warm greenhouse during the preliminary treatment.

Since it had been found in previous experiments that celery did not go to seed under the normal length of day during the short days of winter, especially during December, electric lights were used with half of the plants in each treatment in both houses. The lights were turned on at 5 P. M. and turned off at 10 P. M. Lengthening the day to this extent did not hasten, but rather retarded, the development of the seed stalk of celery. Perhaps the exposure was of too long duration, since there is evidence that the normal daylight during the latter part of February is sufficient for celery. The data secured on the effects of the temperature treatment for periods of 10 to 30

TABLE 2

EFFECT OF TEMPERATURE TREATMENT DURING EARLY GROWTH ON DEVELOPMENT OF SEED STALK IN CELERY

TREATMENT		NUMBER PLANTS	NUMBER OF SEED STALKS AT DIFFERENT DATES			
TEMPERATURE†	LIGHT*		FEB. 26	MAR. 5	APR. 1	MAY 1
C. F. 10 days.....	No light	19	9	10	16	19
C. F. 30 days.....	No light	20	18	18	20	20
C. F. 10 days.....	Light	20	4	11	20	20
C. F. 20 days.....	Light	18	0	5	13	18
C. F. 30 days.....	Light	18	10	17	18	18
Cool house } check...	Light	22	0	4	13	22
Warm house } 30 days..	Light	18	0	0	6	18

†Temperature refers to treatment during early stage of growth. C.F. = cold frame, 10, 20 and 30 days = length of time exposed to preliminary treatment.

\*No light = no artificial light. Light = five hours artificial light from 5 to 10 P. M.

days, during the early stage of growth are given in TABLE 2. After the preliminary treatment, these plants were grown in the cool greenhouse.

All of the plants grown in the cool greenhouse after their preliminary temperature treatment went to seed by May 1, but it is of interest to note that those which had been subjected to high temperature from October 16 to November 15 were delayed more than a month as compared to the others.

The plants kept in the warm house had not developed a single seed stalk by June 1, regardless of the previous treatment. In other words, the high temperature during the main period of growth prevents the development of the seed stalk, even though the plants had previously been subjected to the necessary conditions for reproduction. Some of the plants from the warm house were transferred to the cool house in March and they showed no evidence of going to seed by June 1.

#### LITERATURE CITED

- Starring, C. C., 1924. Premature seeding of celery. *Mont. Agr. Exp. Sta. Bull.* 168.  
 Thompson, H. C., 1924. Factors influencing early development of seed stalk of celery. *Proc. Am. Soc. Hort. Sci.* 1923: 219-224.  
 Whipple, O. B., 1915-1919. Report of Horticultural Department. *Mont. Agr. Exp. Sta. Ann. Rpts.* 21: 396. 1915: 22: 251. 1916: 23: 175. 1917: 24: 253. 1918: 25: 144. 1919.





Relatively high temperatures averaging 70° F. prevent the development of seed stalks of celery. Plants on the left were grown in a cool greenhouse with an average of 60° F., while those on the right were grown in a warm greenhouse with an average of 70° F.





The plants in the center of this view were exposed to relatively low temperatures in a cold frame for five weeks prior to being set in the field. The plants on the adjacent rows are from the same lot of seed but had been grown in a greenhouse under higher temperatures until set in the field.





## STERILITY IN THE COMMON CABBAGE (BRASSICA OLERACEA L.)

L. R. DETJEN

*Delaware Agricultural Experiment Station*

There are a number of kinds of sterility that may be held responsible for the sterility of cabbage plants. Of these, the following may be listed as being of importance: (a) incompatibility; (b) intersexualism; (c) abortion of earliest flower buds; (d) senility; (e) protogyny; (f) protandry, and (g) antholosis. These will be briefly discussed in their order.

*Incompatibility.* Probably the most important type of sterility that is responsible for the unfruitfulness of cabbage plants is that of incompatibility. This phenomenon, because of its variable expressions, is difficult of clear definition and hence only comparative terms will be used to designate its character. Four different types of incompatibilities have been isolated at the Delaware Agricultural Experiment Station and these seem to breed true to type when carefully guarded against cross-pollinations. That these four types of incompatibility differ from one another may be discerned from their general appearance and behavior to pollinations. For our convenience we may designate them by letters of the alphabet and call them *A*, *B*, *C* and *D* respectively.

Type *A* is only feebly self-incompatible and no cross-incompatibility is in evidence except when plants possessing the *C* type are concerned. Plants carrying this *A* type may be described as but slightly self-sterile and fully cross-fertile.

Type *B* is very strongly self-incompatible but no cross-incompatibility is present except where the *C* type is concerned. Plants of this *B* type then are sterile when selfed but fertile when crossed.

Type *C* is highly self-incompatible and equally as highly cross-incompatible within its own hereditary strain. Pollen from other hereditary strains seems equally as ineffective as that from within the strain, but pollen from plants of this type when used to pollinate plants from other strains shows no immediate signs of incompatibility. Thus, this *C* type of incompatibility seems to be intimately associated with the pistils rather than with the pollen of the flower. Plants possessing this *C* type of incompatibility are chiefly self-sterile and cross-sterile within the strain.

Type *D* appears to be much like type *A*, but the self- and cross-incompatibility factors are more pronounced. The divergence of this type from that of *A* becomes evident in the  $F_1$  hybrids with other strains.

Whether incompatibility is hereditary in the Mendelian sense seems to be a matter of controversy. The subject matter is very complicated and too variable in its expressions to permit of hasty conclusions and for this

reason the following briefly recorded observations are submitted with the hope that they might contribute to a fuller understanding of the subject. The evidence here presented is mainly for the purpose of answering the following two questions so often asked by the breeder of plants: (1) Will incompatibility yield to selection? and (2) Can incompatibility be transmitted as such to its progeny by crossing?

The answer to the first question, it seems, is given in the form of five or six strains (now in existence at the Delaware Agricultural Experiment Station) which, through selfing reproduce their kind and carry fairly stable types of incompatibilities.

To answer the second question, "Can incompatibility be transmitted to hybrid progeny as a dominant or recessive?" only parent plants of known incompatibilities were used together with some whose identity has not been fully established. The evidence secured from the hybrids will be presented a little more in detail.

First group of hybrids. A plant of the variety Early Bloomer possessing the *A* type of incompatibility (feebly self- and no cross-incompatibility) was crossed with a plant of the variety Ovate of the *B* type (highly self- but no cross-incompatibility). Reciprocal crosses also were made. Four plants from the progeny, i. e., two of the direct and two of the reciprocal cross, were selected on lines of independent sets of plant characters and set in two rows in the greenhouse for seed production. All four plants gave very uniform results in regard to their incompatibility. All were highly self-incompatible with little or no cross-incompatibility. The *B* type, therefore, was transmitted to all the  $F_1$  plants as a dominant to the *A* type.

Second group of hybrids. A plant of the Ovate variety of the *B* type of incompatibility was crossed with a Zinnia plant of the *C* type (highly self- and cross-incompatible). The reciprocal cross was also made. Four plants were selected from this lot (two direct and two reciprocal) and set in the greenhouse side by side with those of the first group. All four plants gave very uniform evidence, viz., highly self- and cross-incompatibility. In this group the *C* type of incompatibility was transmitted to the  $F_1$  as a dominant to the *B* type.

Third group of hybrids. An Early Bloomer plant of the *A* type was crossed with a Zinnia plant of the *C* type of incompatibility. Again the reciprocal cross was made and two plants of each selection were transferred to the greenhouse. This group was planted with those of the others in double rows. Here again the evidence was clear and uniform; namely, all plants were only very feebly self-incompatible and fully cross-compatible. The *A* type of incompatibility seems to have been transmitted unchanged to the  $F_1$  plants as a dominant to the *C* type.

The following other two lots of hybrids gave similarly uniform and striking evidence, but the type of incompatibility of one or both of the parents is not very clear.

Fourth group of hybrids. A Tall Saucer plant presumably of the *C*

type of incompatibility was crossed with a Flat which was still heterozygous for the *B* and probably *D* types. Only the direct cross was made. Four plants were selected from the  $F_1$  and they also gave very uniform and exceptional evidence. All four plants were highly self- and highly cross-incompatible. Apparently the *C* type was again transmitted from the Tall Saucer parent as a dominant to the other types found in the plant of the Flat selection.

Fifth group of hybrids. The same Tall Saucer plant of the *C* type was crossed with a Narrow Spindle which is to represent the *D* type of incompatibility. The reciprocal cross was again made and four plants from the  $F_1$  were chosen for seed production. These plants together with those of the previous lot were set side by side as a double row on the opposite side of the greenhouse. All four plants were strikingly uniform in the evidence produced. All were highly self-incompatible and medium cross-incompatible like the Narrow Spindle parent. The *D* type thus was not transmitted as such but all hybrids were alike and of a type more resembling that of *B*.

From the evidence secured in these five distinct lots of hybrid plants, it seems logical to conclude that the phenomena of incompatibilities are closely associated with hereditary units and transmitted as allelomorphic pairs to the hybrid progeny.

*Intersexualism.* Intersexualism in plants of *B. oleracea* has been observed regularly for many seasons. Sometimes it is the early flowers that are affected and again at other times almost all of them are so affected. Intersexualism operates to injure or reduce the functions of either one of the sex organs in a flower. Some strains appear to be more affected than are others. The Narrow Spindle strain is one of these. Most of the early flowers are of the type designated as pseudo-staminate. The pistils cease to develop after attaining varying sizes, turn yellow and finally wither away. The stamens continue their normal development and produce an abundance of viable pollen.

The antithesis of the suppression of the pistil produces the pseudo-pistillate blossom, in which the stamens and pollen have ceased development. The pistils in such flowers continue to develop normally and with viable, compatible pollen from other plants set an abundance of seed. Intersexualism seems to affect at times the flowers on the entire plant or only some of them. Later on plants so affected generally return to normal conditions.

*Abortion of the Earliest Flower Buds.* Abortion of those flower buds that are first formed on a seed stalk or its laterals has been observed regularly in *B. oleracea* on all plants when grown either under glass or under normal conditions in the field. The number of buds so affected varies with different plants. Sometimes they are already black or in various stages of distintegration as the heads are opened while at other times they almost reach the blooming stage before death occurs. This phenomenon occurs in the leafy as well as in the head type, and in the more nearly annual types as well as in those that are distinctly biennial in character.

*Senility.* End season sterility occurs frequently in cabbage and seems to be due to senility of the plant. It is manifested at the end of a heavy seed crop.

*Protogyny.\** Protogynous flowers are of frequent occurrence on cruciferous plants. Two types of these have been observed on *B. oleracea*. The most frequent one is of a genetic character resulting in the so-called long-styled flower, while the other type, not so common, seems to be a temporary variation in certain strains resulting in the extrusion of the stigma and frequently the style even from immature flower buds.

*Protandry.* A curious case of protandry has been observed in some strains of cabbage. Kerner and Oliver (1895) state that cruciferous plants are exclusively protogynous. Protandry, therefore, apparently had not been observed in such plants. It may be described as follows: While the flower bud is enlarging the stigma is in some manner prevented from moving forwards with the rest of the pistil. This causes the style to become curved or sometimes doubled over to one side. At time of anthesis the stigma is caught and held for an indefinite period of time by the lip of one of the expanding sepals and actually drawn over to one side of the flower. The anthers shed their pollen in a normal fashion and when finally the stigma is released due to the continued growth of the pistil very little, if any, of the pollen remains for self-pollination. It also appears that the stigma so held becomes receptive at a belated period which fact further interferes with self-pollination of the flower. The number of flowers so affected is variable and the phenomenon lasts for an indefinite period of time. After a while the plants recover and continue in a normal manner.

*Antholosis.* Reversion of carpellary structure and a general return of flower organs to a more vegetative stage has been noticed annually at irregular intervals. Such cases have been reported in other species of *Brassica* by Stout (1922) and need not be here described.

#### LITERATURE CITED

- Kerner, A., and Oliver, F. W., 1895. The Natural History of Plants. 2 vol. New York.  
Stout, A. B., 1922. Cyclic manifestation of sterility in *Brassica pekinensis* and *B. chinensis*. Bot. Gaz. 73: 110-132.



# EFFECT OF SELF-FERTILIZATION IN CABBAGE AND ONION

FELIKS KOTOWSKI

*Institute of Olericulture and Vegetable Breeding  
College of Agriculture, Warsaw, Poland*

Investigations have been carried out in order to determine the deteriorate consequences of self-fertility in cabbage and onion. Two varieties of cabbage—Amager, a Danish late variety, and Warsaw, a Polish early variety—were studied.

No figures as to yield are available for Amager that grew in 1920 and was destroyed through the action of the Polish-Russian war. Nevertheless some interesting data have been gathered and included in the recent report.

Some lateral flower branches were isolated in 1919 on 16 plants of Amager and their productiveness of seed varied very much (Kotowski, 3). The seeds obtained were planted 1920, and for comparison also seeds were sown received from non-isolated branches on the same specimens. The germination took place in cold frames, the seedlings were carefully treated and twice transplanted before set into the open ground. The quality of seed used is expressed as 100 seeds weight, and the weight of fresh matter of seedling in 45 days stage indicates the influence of self-fertility.

TABLE 1  
COMPARATIVE WEIGHTS OF SEEDS AND SEEDLINGS FROM SELF- AND CROSS-FERTILIZATION OF  
AMAGER CABBAGE—1920

NUMBER OF PLANT	SELF-FERTILIZED FLOWERS		CROSS-FERTILIZED FLOWERS		INDEX OF VITAL VIGOR
	WEIGHT OF 100 SEEDS GR.	WEIGHT OF SEEDLING GR.	WEIGHT OF 100 SEEDS GR.	WEIGHT OF SEEDLING GR.	
V.....	0.30	6.8	0.58	6.2	91
VI.....	0.18	4.8	0.65	4.8	100
VII.....	0.48	6.8	....	7.8	115
IX.....	0.55	3.8	0.61	7.5	200
X.....	0.80	3.6	0.70	6.2	172
XI.....	0.74	6.5	0.61	6.	92
XII.....	0.55	5.8	0.59	6.1	118
XIII.....	0.75	5.2	0.62	9.1	175
XIV.....	0.54	7.	0.64	7.3	104
XV.....	0.88	7.9	0.49	6.6	84
XVI.....	0.66	8.2	0.52	7.9	96
Average.....	0.58	5.7	0.60	6.9	122

The variation of vital vigor, given as percentage of cross-fertile seedling weight in self-fertile, was quite ample, that fact may be referred to the genetic differences of plants in regard to self-fertility (Kotowski, 3, also the remarks in Jones, 2). In average the superior seedlings were those of cross-fertilized

flowers, however they prevail, over self-fertilized but little, i. e., 22 per cent of fresh weight, the quality of seeds being the same. It seems from the data cited there was a very moderate effect of self-fertility in this variety of cabbage, as far as concerning the youth of the plant. In the seedlings given there was no influence of seed weight on the developed seedling. Measurements of correlation between these characters, applying the calculation method of Pearson (6), determined the coefficient  $r = + 0.07$  for self-fertile, and  $r = + 0.20$  for cross-fertile seeds. There was no reason for supposing that such a case will occur in general in cabbage and therefore an additional observation has been made in 1925, planting under field conditions the self- and cross-fertilized seeds of the variety Warsaw.

The seeds belonging to six strains of this variety were sown separately for each strain in cold frames and reaching 60 days stage were transplanted into the field. The treatment was alike for self- and cross-fertilized seeds. The seeds were graded on screens to insure equal size and hence uniform

TABLE 2  
WEIGHTS OF SEEDS AND SEEDLINGS DERIVED FROM SELFING AND CROSSING IN  
WARSAW CABBAGE

NO. OF STRAIN	SELF-FERTILIZATION		CROSS-FERTILIZATION		INDEX OF VITAL VIGOR OF SEEDLING
	HUNDRED SEEDS WEIGHT GR.	WEIGHT OF SEEDLING GR.	HUNDRED SEEDS WEIGHT GR.	WEIGHT OF SEEDLING GR.	
1.....	0.59	1.12	0.60	1.63	146
2.....	0.46	1.41	0.54	1.73	123
3.....	0.49	1.70	0.52	1.95	115
4.....	0.69	1.71	0.73	2.80	164
5.....	0.35	1.90	0.47	2.61	137
6.....	0.57	1.00	0.56	1.75	175
Average.....	0.53	1.47	0.57	2.08	142

TABLE 3  
WEIGHT OF FULLY DEVELOPED HEADS OF SELFED AND CROSSED SEED OF WARSAW CABBAGE

NO. OF STRAIN	SELF-FERTILE FLOWERS					CROSS-FERTILE FLOWERS					INDEX OF VITAL VIGOR (HEAD ED PLANT)	INDEX OF VITAL VIGOR (NON- HEAD- ED PLANT)
	PLANTS HEAD- ED	PLANTS NON- HEAD- ED	PLANTS LOST	WGT. OF HEAD- ED PLANT KG.	WGT. OF NON- HEAD- ED PLANT KG.	PLANTS HEAD- ED	PLANTS NON- HEAD- ED	PLANTS LOST	WGT. OF HEAD- ED PLANT KG.	WGT. OF NON- HEAD- ED PLANT KG.		
1	70	37	10	1.11	1.21	69	39	12	1.73	1.35	156	112
2	52	33	16	2.43	1.47	50	48	10	2.34	1.48	97	102
3	41	39	16	2.25	1.25	21	32	12	2.10	1.33	93	107
4	21	35	13	1.99	1.25	81	38	7	3.10	2.48	156	198
5	56	18	6	3.03	1.92	47	31	8	2.47	1.39	82	73
6	62	63	43	1.20	0.87	78	34	8	2.07	1.43	173	163
	Σ372	Σ225	Σ104	av.2.00	av.1.33	Σ416	Σ222	Σ57	av.2.30	av.1.58	av.126	av.126

seeds from 2 to 2.5 mm. diameter were used. The plants were set 50x50 cm. and grew on sandy but fertile soil. The harvesting was done October 1, and the weighing of plants immediately took place. It may be mentioned that every seedling was marked in the spring and the developed plant received the same number in the fall. This manner of description allowed to emphasize the individual growth of each of the seedlings grown. TABLE 2 and TABLE 3 show the details of the appreciation of Warsaw cabbage.

The seedling weight in 1925 is much smaller than that in 1920. The index of vital vigor is greater in the latter case and it is expressed as 142 against 122 stated for Amager.

It is probable this fact might be due to better vegetation conditions, which supported to the smoothness of differences in the two groups of Amager variety contrary to the Warsaw cabbage. This conclusion is of practical value, because such a behavior of plants observed can give a helpful method in cabbage breeding, diminishing the risk of obtaining weak seedlings from selfed plants.

The estimation of adult plants in the fall is presented on TABLE 3. The influence of cabbage selfing varies in accord to the strain planted. The heading process was not dependent on seed origin, that is evident from the ratios: headed and non-headed plants, which was 150:100 and 146:100. The greater hardness of plants from cross-fertilized flowers was observed, 15 per cent versus 8 per cent. The indices of vital vigor show the effect of cross-fertility as the increase of 26 per cent of weight which occurred the same in headed and non-headed plants.

The inter-relations between the weight of the seedling and the heading process have been stated by means of correlation coefficients (Pearson, 5).

There may be failure of forming heads in cabbage when the weight of seedling diminishes regardless of the seed origin. The same occurred owing to the weight of the head and the weight of the seedling.

If comparison is made between indices of vital vigor of the seedlings and those of the adult plants, we meet minor indices but little in the later stage of cabbage: 146 and 126. It is probable that inbreeding phenomena in cabbage are more limited in further plant development.

TABLE 4  
COEFFICIENTS OF CORRELATIONS, ( $r$ ) FOR THE WARSAW CABBAGE

NUMBER OF STRAIN	HEADING PROCESS AND WEIGHT OF SEEDLING		WEIGHT OF HEAD AND WEIGHT OF SEEDLING	
	SELF-FERTILE FLOWERS	CROSS-FERTILE FLOWERS	SELF-FERTILE FLOWERS	CROSS-FERTILE FLOWERS
1.....	+.107	-.247	+.118	+.097
2.....	-.411	-.490	+.588	+.540
3.....	-.722	-.332	+.234	+.328
4.....	-.490	-.298	+.333	+.203
5.....	-.506	-.310	+.401	+.184
6.....	-.322	-.255	+.251	+.403

The second plant studied was onion, variety Zittauer. The isolation and yield of seed are presented in a former publication (Kotowski, 4). TABLE 5 shows the data for 26 strains of onion. Their vital vigor is expressed as above. The increasing of vigor in favor of crossed plants was slight (14 per cent) and some of the strains show no differences between selfed and crossed progenies.

TABLE 5  
DATA FOR THE ZITTAUER ONION

NO. OF STRAIN	WEIGHT OF BULB IN GRAMS		INDEX OF VITAL VIGOR	NO. OF STRAIN	WEIGHT OF BULB IN GRAMS		INDEX OF VITAL VIGOR
	SELFED PLANTS	CROSSED PLANTS			SELFED PLANTS	CROSSED PLANTS	
9	140	163	117	72	174	175	100
13	145	175	121	80	122	114	93
15	126	159	125	84	130	134	103
17	175	160	92	86	125	136	109
23	128	138	109	90	116	114	99
27	120	183	154	95	69	131	189
32	100	126	127	96	114	110	97
37	126	169	133	107	103	101	99
39	149	123	82	109	77	92	119
45	200	226	112	115	63	79	125
54	142	138	97	118	70	88	125
57	117	142	118	129	104	97	93
60	122	176	143	130	80	96	121
				Average. . .	120	137	114

### CONCLUSIONS

The data gathered hereby seem to justify the conclusion that in breeding work with cabbages and onions the isolation process is a valuable method.

The phenomena of constitutional weakness, as a consequence of this method, exist only in a moderate degree and therefore the inbred progenies may be a worthy material for further breeding investigations.

### LITERATURE CITED

1. East, E., and Jones, D. F., 1919. Inbreeding and outbreeding.
2. Jones, D. F., 1925. Genetics.
3. Kotowski, F., 1921. The effect of self-fertilization on cabbage. Mem. Inst. Nat. Polon. d'Econom. rurale Pulawy. T. I.
4. ——— 1926. The efficiency of self- and cross-fertility in the onion. Acta. Soc. Botan. Poloniae IV. 1926.
5. Pearson, K., 1909. On a new method of determining correlation between a measured character A, and a character B, of which only the percentage of cases where in B exceeds (or falls short of) a given intensity is recorded for each grade of A. Biometrika 7: 96-105.
6. ——— 1907. On further methods of determining correlations. Biometric Series 4.



## SELF-STERILITY OR FERTILITY IN TRIFOLIUM AND MELILOTUS

A. J. PIETERS

*Bureau of Plant Industry, U. S. Department of Agriculture*

In common with many other members of the order Leguminosae, the flowers of species of *Trifolium* and of *Melilotus* appear to be peculiarly adapted to pollination by insects. That such structures do not, however, necessarily imply self-sterility, is clear from such a species as the sweet pea, which is self-fertile and does not commonly cross in the field. Of all species of *Trifolium*, the red clover, *T. pratense*, is the most important and, since Darwin pointed out the relation between bumble bees and the red clover seed crop, there has been much interest in the question of whether red clover is or is not self-sterile. No attempt will be made in this brief presentation of the essential evidence to refer to all the literature. This has been adequately done by those workers to whom reference will be made, more especially by Williams, at the University College of Wales, Aberystwyth.

Further, most of this older work is at present of historical interest only since the methods employed were crude or inadequate. It was evident, however, that seeds were sometimes produced after self-pollination, and in 1883 Armstrong suggested that, while red clover is in the main self-sterile, there may be individual plants capable of being self-fertilized. Evidence for this view was secured by Fergus in 1922, and later additional evidence was presented by Kirk in 1925, and again by Williams in 1925. These workers have shown that, when pollen from a given red clover plant is placed on a stigma of the same plant fertilization may take place, though most of the self-pollinated flowers failed to produce any seed whatever. Fergus self-pollinated one head on each of 650 plants and secured some seed from 32 plants or from about 5 per cent of the plants studied. Williams secured seed from 30 per cent of plants self-pollinated. Each of these workers found, however, that in most cases very few seeds were produced. Out of 32 self-pollinated plants from which Fergus secured seed, 21 yielded less than 5 seeds per plant, while only 5 yielded more than 10. Williams found a wide variation ranging from complete sterility to the production of 23.5 seeds per 100 selfed flowers.

Thanks to the three workers mentioned, our knowledge of self-fertility in red clover has become more definite and it is possible to say that some red clover plants are more or less self-fertile, though the probable percentage of such plants in any given population cannot at present be stated. In this connection it must be pointed out that the securing of self-pollination is of importance. In the undisturbed red clover flower the anthers are situated below the stigma, and it is impossible for the pollen from a given flower to

reach the stigma of that flower. Some mechanical means of bringing the pollen to the stigma is therefore necessary, and gentle rolling of the flower head between the thumb and fingers has been found effective. It seems probable that Darwin's failure to obtain any seed from bagged heads was due to lack of self-pollination.

In this connection it is pertinent to call attention to the fact that self-pollination is a very different phenomenon from self-fertility. The placing of the pollen of a given flower on the stigma of that flower is a mechanical process and the floral structure of a flower may be such as to render such transfer difficult or impossible, but when the transfer is effected fertile seeds may be produced. On the other hand, as has been shown to be the case in most red clover plants, the pollen of a given flower may not find a congenial home on a stigma of a flower from the same plant and, because of this physiological inhibition, may fail to fertilize the ovule. In this case self-sterility results.

It is also possible that further studies may show a greater potential self-fertility than can at present be assumed. Williams found that, when red clover flowers were self-pollinated in the bud stage, a higher percentage of seed was set than when flowers were selfed in early or in late bloom, and he suggests that this fact may be correlated with the slow growth of the pollen tube in selfed flowers.

Westgate, Coe and Martin found that in selfed flowers of red clover the growth of the pollen tube was abnormally slow so that the ovule had disintegrated before the tube reached the ovule. By selfing in the bud stage, however, more time is allowed for the growth of the pollen tube. From present information, therefore, it seems possible that self-sterility in red clover may be due to slow growth of the pollen tube in selfed flowers rather than to any physiological repulsion between pollen and ovule nuclei.

Fergus, however, believes that the capacity for self-fertility is an inherited character and has presented evidence to show that some of the descendants of plants presenting a high degree of self-fertility also show a high degree of self-fertility. From the standpoint of establishing pure lines of red clover this feature is naturally of importance, but more work will be necessary before definite conclusions can be drawn.

While, therefore, there remains no doubt that red clover seed may be produced as a result of self-fertilization, it is certain that seed produced in the open field is prevailingly, if not nearly always, the result of cross-pollination, and this fact must be reckoned with in considering the possibility of producing and maintaining improved varieties.

In regard to other species of the genus *Trifolium*, very little work has been done as far as the writer knows. Kerner von Marilaun indeed makes the broad statement that most species of *Trifolium* ripen seed when unvisited by insects. Unfortunately, the writer has not been able to search the literature for evidence of the above statement. The limited experience which the writer has had with growing species of *Trifolium* in the greenhouses of the

U. S. Department of Agriculture tends to the opposite conclusion. *Trifolium incarnatum* is believed to be highly self-fertile, but the present writer is not aware of any definite evidence on this point. At the U. S. Department of Agriculture attempts have been made to obtain seed from individually caged plants of *T. repens* and of *T. hybridum*, but without success. In view of the work on red clover discussed above, this work needs to be repeated with different technique. One species, *T. multinerve* (Hochst.) A. Rich., an Abyssinian species, is definitely self-fertile, as single plants raised in the greenhouses of the Department of Agriculture have set seed freely. There was in this case no possibility of cross-pollination, as there was but one plant of the species at a time.

*Melilotus*. The results secured by workers on species of *Melilotus* are not so much at variance as those already reported for red clover. Some of the older workers studied *Melilotus alba*, while others worked with *Melilotus officinalis*. Darwin found that *M. officinalis* was self-pollinated to a small extent only, and Knuth gives this species as an example of self-sterility. These findings have more recently been confirmed by Kirk, at Saskatoon, and it may therefore be concluded that *M. officinalis* is usually self-sterile. However, there is nothing in Kirk's work to show that any attempt was made to assure artificial self-pollination and it is not impossible that, as with red clover, the chief difficulty may lie with self-pollination rather than with self-fertility.

Coe and Martin, in the United States, and Kirk, in Canada, published the results of work with *Melilotus alba* and in this case the results vary somewhat. Coe and Martin bagged certain racemes, left others exposed, and artificially self- or cross-pollinated still others. They found that protected, undisturbed flowers set seed but seldom, while flowers that had been rubbed between the fingers or had been artificially self-pollinated set seed quite freely, though not so freely as exposed flowers or as those artificially cross-pollinated. It is evident that in the bagged flowers self-pollination did not often occur, but that when self-pollinated the flowers of *M. alba* were highly self-fertile.

Kirk found that the average number of pods per raceme secured from caged plants of *Melilotus alba* was a trifle more than half that secured from unprotected plants, and he also secured a fair number of pods from bagged racemes. The disagreement between the work of Coe and Martin and that of Kirk pertains, however, to self-pollination rather than to self-fertilization. In each case it was found that *Melilotus alba* is highly self-fertile, provided self-pollination is assured. Why self-pollination should be more readily secured in Saskatoon, Saskatchewan, where Kirk worked, than at Ames, Iowa, or at Arlington, Virginia, where the work of Coe and Martin was done, cannot be definitely stated, but the present writer may suggest a greater movement of air currents or wind velocity in the one case than in the other. In some selection work which the writer did on annual white sweet clover, it was found that caged plants set seed freely when the cage, after being well filled by the growing and blossoming plant, was violently shaken from time

to time. On a dry, clear day it was possible on shaking the cages to note considerable quantities of pollen in the air, showing that the mechanical disturbance had affected the ripe anthers.

The information with regard to *Melilotus alba* may, therefore, be summarized as showing definitely that this species is highly self-fertile and that the degree to which self-pollination occurs may vary with conditions. The evidence at hand does not permit of a positive statement as to whether seed normally produced in the open field is the result of self or of cross-pollination, either one may occur, but the fact that a dwarf variety, as the Grundy County, retains its varietal characteristics, would seem to indicate that self-fertilization is the rule rather than the exception in this species.

*Melilotus officinalis* is certainly prevailingly self-sterile, though work on artificial self-pollination needs to be done before final conclusions can be reached.

Other species of this genus have not been studied.

#### LITERATURE CITED

- Coe, H. S., and Martin, J. N., 1920. Sweet clover seed. I. Pollination studies of seed production. II. Structure and chemical nature of the seed coat and its relation to impermeable seeds of sweet clover. U. S. Department of Agriculture Bulletin 844.
- Fergus, Ernest, 1922. Self-fertility in red clover. Ky. Agr. Exp. Sta. Cir. 29.
- Kirk, Lawrence E., 1925. Artificial self-pollination in red clover. Sci. Agr. 5: 179-189.
- 1925. Self-pollination of sweet clover. Sci. Agr. 6: 109-112.
- Westgate, J. M., and Coe, H. S., 1915. Red clover seed production. U. S. Dept. Agr. Bulletin 289.
- Williams, R. D., 1925. Studies concerning the pollination, fertilization and breeding of red clover. University College of Wales, Welsh Plant Breeding Station, Series H, No. 4.



# TYPES OF STERILITY IN WILD AND CULTIVATED POTATOES

CHARLES F. CLARK

*Bureau of Plant Industry, U. S. Department of Agriculture*

The tuber-bearing species of the genus *Solanum*, considered as a group, cover a wide range of distribution in the Western Hemisphere, the cultivated varieties, known botanically as *S. tuberosum*, being grown to a greater or less extent from Southern Alaska and the Canadian provinces to Southern Chile, while the range of the wild species extends from the southwestern part of the United States to the southern part of South America. It is therefore not surprising to find considerable diversity of type with respect to certain characteristics of this group of plants, including not only modifications of the vegetative parts, but of the reproductive organs as well, which influence in various ways the fruitfulness of the plant.

The material on which these studies are based consisted of 170 commercial varieties and 513 new seedling varieties of the cultivated potato (*Solanum tuberosum* L.) from several different countries and 9 wild species and hybrids, all of which were grown at Presque Isle, Maine. Certain of these were also grown at the New York Botanical Garden. The behavior of many of these varieties and species with respect to sterility has previously been reported by Stout and Clark (8). The following types of sterility have been distinguished: (1) Sterility due to premature abscission of buds and flowers; (2) sterility from one-sided impotence, or intersexualism; (3) Sterility due to hybridity, and (4) Sterility resulting from physiological incompatibility.

## STERILITY DUE TO PREMATURE ABSCISSION OF BUDS AND FLOWERS

The early shedding of buds or flowers is more or less strongly developed in the cultivated varieties but has not been observed to any extent in any of the wild species which have been included in the author's investigations. East (4) concluded from a study of 721 varieties that there are all gradations of this condition, including those varieties whose buds drop off without opening, those in which a few flowers open but immediately fall, and those whose flowers persist for several days. Cytological studies by Young (9) have shown that, coincident with the process of early abscission and the accompanying withering of the bud or flower, there is a degeneration of both ovules and anther contents.

Premature abscission constitutes a very effective type of sterility since it is obvious that buds which fall before opening or flowers which persist for only a few hours cannot produce fruit. Where the expression of this condition is not very pronounced, so that a few flowers open and persist for a few

days, they may under favorable climatic conditions produce fruit when pollinated with viable pollen. The anthers of such flowers produce but little pollen, which is rarely if ever viable.

While the degree of non-blooming varies with the variety, it is also influenced by conditions of environment. This is clearly shown by an experiment conducted by Stout and Clark (8) in which halved tubers of 15 varieties were grown, one set at Presque Isle, Maine, the other, consisting of the corresponding halves, at the New York Botanical Garden. All the varieties bloomed profusely at Presque Isle. At the New York Botanical Garden only two of the same varieties bloomed well, three produced a few flowers, while ten produced no flowers which opened.

This is apparently a physiological phenomenon varying with the variety in the intensity of its response to environmental conditions.

### STERILITY FROM ONE-SIDED IMPOTENCE, OR INTERSEXUALISM

In the cultivated varieties of *Solanum tuberosum* pollen sterility is very strongly manifested. This condition appears to be inherent in the species, occurring in all varieties which have come under the author's observation. Studies of the pollen of the 170 commercial and 513 seedling varieties previously referred to, representing material from many parts of the world, have failed to show a single variety in which there was not a fairly large percentage of sterile pollen grains. Of the seven wild species studied, including *S. Caldasii* var. *glabrescens* Dunal, *S. chacoense* Bitt., *S. Commersonii* Dunal, *S. demissum* Lindl., *S. Fendleri* Gray, *S. Jamesii* Torr., and *S. polyadenium* Greenm., only one, *S. Commersonii*, showed the presence of this type of sterility in any marked degree. So far as investigations have shown, the intersexual type of sterility in the potato involves only the male organs. The results of several hundred crosses between a large number of different varieties have given no evidence of female sterility.

This sterility is the result of degeneration of the pollen grains. Breeze (2) has distinguished 3 conditions of pollen degeneracy in the potato: (1) shriveled and empty pollen grains; (2) hypertrophied or swollen grains; and (3) absence of pollen grains. The deterioration resulting in shriveled and empty pollen grains was found to occur after the formation of the pollen mother cell. The breaking down of the pollen grains at a comparatively late stage in their development is further confirmed by Dorsey (3), who concludes that the development of the pollen grains is stopped after they are liberated from the tetrad, and by Young (9), who states that their disintegration occurs when they are nearly mature.

A grouping of potato varieties into 4 classes, based on the condition of the anthers, the quantity of pollen shed, the relative number of grains possessing granular contents, and the extent and vigor of germination of pollen, has been made by Stout and Clark (8), as follows:

1. Anthers well developed, orange colored; pollen abundant, at least 30 per cent of grains plump; germination good, 15-20 per cent germinating.

2. Anthers well developed, orange colored; pollen fairly abundant, percentage of plump pollen low (5 per cent to less than 3 per cent), few grains germinating, tubes of feeble growth.

3. Anthers usually well developed, occasionally somewhat greenish at apex; pollen scant, usually less than 10 per cent and often less than 1 per cent of grains plump; rarely any germination.

4. Anthers mostly greenish; dehiscence irregular; pollen very scant with scarcely a plump grain or pollen grains lacking; no germination.

### STERILITY DUE TO HYBRIDITY

The data bearing on the occurrence of this type of sterility in the potato are rather meager since only three hybrid forms have been available for study.

The original stock of the first of these was obtained from Lima, Peru, under the name of *Solanum Maglia*. This is, however, not the true *S. Maglia* Schlecht. By crossing with *S. chacoense*, which breeding experiments have shown to be a pure species, a few plants were obtained for genetic studies. The wide variations, including distinct types, in the  $F_1$  progeny indicate very clearly that the form in question was of hybrid origin. Further confirmation of this is afforded by the cytological studies of H. B. Smith (unpublished work) who reports that the chromosome behavior, as determined from preliminary observations by Belling's method, was found to correspond to that of known hybrids. Examination of the pollen showed that at least 95 per cent of the grains were empty and shriveled. To determine the ability of the female organs to function, crosses were made, using *S. Caldasii*, var. *glabrescens*, *S. chacoense*, *S. demissum*, *S. Fendleri*, *S. Jamesii*, and *S. tuberosum* (var. "McCormick"), respectively, as the pollen parents. No fruits resulted from the pollinations of the four species last named. Fruits were obtained from the *S. Caldasii* var. *glabrescens* crosses which contained a few seeds, none of which germinated. The *S. chacoense* crosses resulted in the production of a few fruits which contained seeds from which a few  $F_1$  plants were obtained. Also sixty-eight flowers were selfed, none of which produced fruits. It thus appears that partial female as well as male sterility occurs in this form.

The second hybrid studied was one which resulted from a cross between *Solanum Fendleri* and *S. chacoense*. The pollen of the plants of this hybrid contained very few plump grains. Flowers pollinated with their own pollen or with pollen of the female parent, *S. Fendleri*, did not produce seed balls but small fruits containing no seeds, which dropped off before reaching full size, were produced in many instances when *S. chacoense* was used as the pollen parent. In this hybrid there was almost complete sterility of both male and female organs.

An examination of the pollen of the third hybrid, which was a cross between *Solanum demissum* and *S. chacoense*, showed the presence of but very few plump grains. No data were obtained bearing on the question of female sterility in this hybrid.

The male sterility exhibited in these hybrids appears to be identical with that of the most extreme cases of intersexualism observed in the cultivated varieties of *Solanum tuberosum*. In the hybrids, however, both maleness and femaleness are involved. This appears to be characteristic of sterility resulting from hybridity. Further confirmation of this distinctive feature of hybrid sterility is seen in the hybrids between members of certain other groups of plants. In hybrids obtained by crossing *Digitalis lutea* and *D. purpurea*, Haase-Bessel (6) found abortion of both the pollen grains and egg cells. Cytological studies of hybrids between radish and cabbage, by Karpechenko (7), and breeding experiments by Gravatt (5), with hybrids of similar origin, showed the presence of both male and female sterility. Belling (1) reports semi-sterility in both male and female organs of hybrids obtained by crossing the Florida Velvet bean (*Stizolobium Deeringianum*) with the Lyon bean (*S. niveum*), the Yokohama bean (*S. Hassjoo*), and the China bean (*S. niveum* var.), respectively, in which one-half of the pollen grains were empty and one-half the ovules had no embryo sacs.

### STERILITY RESULTING FROM PHYSIOLOGICAL INCOMPATIBILITY

In connection with the potato breeding experiments of the United States Department of Agriculture at Presque Isle, Maine, many hundreds of con-

TABLE 1  
RESULTS OF HAND POLLINATIONS IN *Solanum Caldasii* VAR. *glabrescens*, 1923-1924  
SELFED

PLANT	NUMBER OF	
	FLOWERS POLLINATED	SEED BALLS
No. 6.....	9	0
No. 15.....	14	0
No. 16.....	3	0
No. 17.....	10	0
No. 23.....	7	0
No. 24.....	2	0

### CROSSES BETWEEN DIFFERENT PLANTS WITHIN THE SPECIES

PARENTS BY NUMBER	NUMBER OF	
	FLOWERS CROSSED	SEED BALLS
1 x 6.....	3	2
2 x 6.....	4	0
3 x 6.....	11	0
4 x 6.....	8	8
5 x 6.....	5	0
8 x 6.....	8	0
8 x 10.....	9	0
10 x 6.....	6	0
13 x 6.....	5	0
14 x 10.....	6	1
		3



TABLE 2  
RESULTS OF HAND POLLINATIONS IN *Solanum chacoense*, 1921-1924  
SELFED

PLANT	NUMBER OF		PLANT	NUMBER OF	
	FLOWERS POLLINATED	SEED BALLS		FLOWERS POLLINATED	SEED BALLS
No. 1.....	8	0	No. 25.....	2	0
No. 2.....	17	0	No. 26.....	3	0
No. 3.....	8	0	No. 27.....	4	0
No. 6.....	6	0	No. 29.....	7	0
No. 7.....	4	0	No. 38.....	3	0
No. 8.....	7	0	No. 39.....	2	0
No. 10.....	5	0	No. 40.....	7	0
No. 12.....	8	0	No. 43.....	4	0
No. 13.....	9	0	No. 44.....	7	0
No. 15.....	7	0	No. 49.....	5	0
No. 18.....	9	0	No. 50.....	14	0
No. 19.....	5	0	No. 51.....	17	0
No. 21.....	8	0	No. 52.....	3	0
No. 24.....	9	0			

## CROSSES BETWEEN DIFFERENT PLANTS WITHIN THE SPECIES

PARENTS BY NUMBER	NUMBER OF		PARENTS BY NUMBER	NUMBER OF	
	FLOWERS CROSSED	SEED BALLS		FLOWERS CROSSED	SEED BALLS
1 x 2.....	7	0	21 x 29.....	9	0
1 x 3.....	9	0	21 x 40.....	8	0
1 x 5.....	8	0	22 x 26.....	2	0
2 x 5.....	6	0	26 x 3.....	6	0
2 x 18.....	1	0	26 x 18.....	9	0
3 x 1.....	8	0	26 x 29.....	9	0
3 x 5.....	9	0	26 x 43.....	9	0
3 x 8.....	2	0	26 x 50.....	3	0
3 x 13.....	6	0	29 x 1.....	4	0
3 x 18.....	7	0	29 x 2.....	5	0
3 x 21.....	7	0	29 x 18.....	26	5
3 x 40.....	4	0	29 x 21.....	9	0
3 x 50.....	5	0	29 x 26.....	8	0
4 x 21.....	10	10	29 x 50.....	9	0
5 x 3.....	7	0	40 x 21.....	10	0
5 x 18.....	4	0	40 x 29.....	9	0
6 x 2.....	4	0	40 x 43.....	8	0
6 x 26.....	5	0	40 x 50.....	5	0
7 x 2.....	7	0	40 x 51.....	6	0
7 x 21.....	6	0	43 x 26.....	7	0
8 x 3.....	5	0	43 x 40.....	4	0
8 x 18.....	4	0	44 x 18.....	9	0
9 x 5.....	6	0	44 x 29.....	7	0
10 x 3.....	10	0	45 x 3.....	4	0
10 x 8.....	5	0	47 x 21.....	6	6
10 x 40.....	8	0	47 x 29.....	2	2
11 x 5.....	2	0	49 x 50.....	7	7
13 x 8.....	13	1	50 x 2.....	1	0
13 x 8.....	23	0	50 x 3.....	7	0
13 x 11.....	1	0	50 x 5.....	8	0
13 x 18.....	3	0	50 x 8.....	9	0
13 x 29.....	6	0	50 x 10.....	14	0
13 x 50.....	7	0	50 x 18.....	2	0
14 x 5.....	2	0	50 x 21.....	13	0
17 x 21.....	2	0	50 x 26.....	6	0
18 x 1.....	12	0	50 x 29.....	5	0
18 x 2.....	5	0	50 x 40.....	6	0
18 x 21.....	21	2	50 x 44.....	4	0
18 x 29.....	10	0	50 x 49.....	7	3
18 x 150.....	12	5	51 x 5.....	10	0
20 x 18.....	6	6	51 x 26.....	2	0
21 x 3.....	5	0	51 x 40.....	5	0
21 x 13.....	10	0	150 x 18.....	8	6
21 x 18.....	8	0			

trolled pollinations have been made, involving not only the cultivated varieties of *Solanum tuberosum* but the wild species previously enumerated. The results of these pollinations have disclosed the presence of a type of sterility known as physiological incompatibility which was found to be very highly developed in two species, *S. Caldasii* var. *glabrescens* and *S. chacoense*. The records for the pollinations in these species, made during the years 1921 to 1924, inclusive, are given in Tables I and II. In the material under investigation the plants of both species produced pollen with nearly 100 per cent perfect grains which gave a relatively high percentage of germination on artificial media, nevertheless, not one of these plants set fruit to its own pollen and only 40 per cent of the cross-pollinations within the species *S. Caldasii* var. *glabrescens* and 12.8 per cent in *S. chacoense* produced seed balls. Certain combinations, however, were found in which fruit set abundantly.

Of the other species studied, *Solanum demissum*, *S. Fendleri*, and *S. polyadenium* are highly self- and cross-compatible; *S. Jamesii* appears to be somewhat feebly self- and cross-compatible, at least under the conditions which prevail at Presque Isle, Maine, while the compatibility relationships of *S. Commersonii* are not apparent because of the predominance of pollen abortion. In the varieties of *S. tuberosum* no well defined instances of physiological incompatibility have been found though there are indications of some slight variations in the degree of compatibility.

### SUMMARY

In the material studied, which consisted of cultivated varieties, wild species and species hybrids, belonging to the tuber-bearing section of the genus *Solanum*, four types of sterility were found: (1) Inhibition of fruitfulness resulting from premature abscission of buds and flowers, occurring in *Solanum tuberosum*. (2) Pollen abortion, or intersexualism, present in all varieties of *S. tuberosum*. Pollen sterility, probably of this type, is also present in *S. Commersonii*. (3) Sterility of hybrids involving both male and female organs. (4) Physiological incompatibility causing complete self-sterility, also cross-sterility except in certain combinations of crosses. This type occurs in *S. Caldasii* var. *glabrescens* and in *S. chacoense*.

### LITERATURE CITED

1. Belling, John, 1914. A study of semi-sterility. Jour. Heredity 5: 65-73.
2. Breeze, Mable S. G., 1921. Degeneration in anthers of potato. Gard. Chron. Ser. 3, 70: 274-275.
3. Dorsey, M. J., 1919. A note on the dropping of flowers in the potato. Jour. Heredity 10: 226-228.
4. East, Edward M., 1908. Some essential points in potato breeding. Conn. Agr. Exp. Sta. 31st/32nd Ann. Rept. (Bien. Rept.), 1907/1908: 429-447.
5. Gravatt, Flippo, 1914. A radish-cabbage hybrid. Jour. Heredity 5: 269-272.
6. Haase-Bessel, G., 1916. Digitalis-Studien I. Zeitschr. f. induct. Abstammungs- u. Vererbungslehre 16: 293-314.
7. Karpechenko, G. D., 1924. Hybrids of *Raphanus sativus* L.  $\times$  *Brassica oleracea* L. Jour. Genetics 14: 375-396.
8. Stout, A. B., and Clark, C. F., 1924. Sterilities of wild and cultivated potatoes with reference to breeding from seed. U. S. Dept. Agr. Bull. 1195.
9. Young, W. J., 1922. Some phases of breeding work and seed production of Irish potatoes. S. C. Agr. Exp. Sta. Bull. 210.

# MICRO AND MACROSPOROGENESIS IN SUGAR BEET WITH SPECIAL REFERENCE TO THE PROBLEM OF INCOMPATIBILITY

ERNST ARTSCHWAGER

*Bureau of Plant Industry, U. S. Department of Agriculture*

The question of self-sterility and inbreeding in beets has been a vital one throughout the history of beet culture, and it is an open one even today.

Early workers obtained both positive and negative results in their breeding experiments, and the same may be said of later experimenters who worked under more rigorously controlled conditions. The plants were either grown in isolated places, far removed from other beets, or they were enclosed in cloth cages to keep out foreign pollen. But since it was shown that even the best cloth would let pollen pass through, Nillson, in his experiments, enclosed branches of the inflorescence axis in suitable paper bags. He obtained positive results and a varying degree of self-fertility which he claims is inherited. Dudok van Heel also obtained positive results and with Nillson he believes that there is self-fertility in beets but that like in rye there appear to be self-sterile and self-fertile races.

In my own experiments I used paper bags, and for the isolation of flower cluster or individual flowers the following device: one-half of a celluloid capsule such as can be obtained in any drugstore was placed over a more or less isolated flower cluster; it was secured in position by means of a small amount of plastic clay. By this method and by the use of paper bags I obtained a number of well-developed seeds.

Since it is an accepted belief, though the facts have never been carefully investigated, that the beet flowers are distinctly protandrous, it should first of all be ascertained whether close-pollination is theoretically possible or whether at best only geitonogamy exists. A study of the development of the pollen and embryo sac shows that the anthers begin development earlier than the ovule and that the former precede the latter stage for stage. Meiosis in the pollen mother cells takes place before the functional megaspore is determined and the pollen is mature before the three divisions in the embryo sac are completed. However, when the flowers open and the pollen is actually shed, the embryo sac contains its eight nuclei in their relative position and theoretically the egg should be ready to be fertilized.

Since it will be needless to enter into details of the nuclear phenomena in the development of pollen and embryo sac, only a short summary will be given with emphasis on the points which have a direct bearing on the problem under discussion.

The stigmatic lobes of the flower begin to expand several hours or even half a day after the flower opens and sheds its pollen. This observation

differs from those of earlier workers according to whom the lobes expand only several days after the opening of the flower. Dudok van Heel, however, in his recent publication, supports the contentions of the writer. He states that when the flowers begin to open in the morning the stigmatic lobes also gradually expand ready to receive the pollen and that even before the lobes have expanded, the pollen tube of the germinating pollen will grow down the stylar canal and affect fertilization and that therefore the flowers could not be protandrous. The egg, then, according to Dudok van Heel, will be fertilized the day when the flower opens, but since the embryo does not start development before the third or fourth day after the opening of the flower, the fertilized egg is supposed to undergo a period of rest. The writer's own observations, however, limited because of lack of material yet positive in that they are based on more critical stages, indicate that the egg is very likely not fertilized the same day and that the egg once fertilized goes on dividing after experiencing only a limited period of rest or none at all.

However the case may be, it should not alter the fact that self-fertilization is a theoretical possibility which has been proved experimentally by Briem, Fruwirth and in a few instances by myself. Self-fertility or at least geitonogamy is to all appearances possible, but we can not, nevertheless, get away from the fact that in isolated plants the production of seed is much smaller and that the seed obtained is apt to be sterile or produce less vigorous progeny than cross-fertilized plants. What are the causes of such a behavior?

A close study of the nuclear phenomena in pollen formation shows that sporogenesis in the anther of the beet offers no noteworthy departure from other plants. The appearance of the chromosomes in diakinesis is regular and there are no unpaired univalents as far as could be determined. The nuclear plate in the heterotypic anaphase contains 9 chromosomes. This number was found to be typical for 20 selected strains of cultivated beets which were studied by the writer. Occasionally a larger or smaller number was observed but such irregularities occur also in other plants and should have no general significance. The anaphasic separation is also quite normal, though occasionally there is a lagging of chromosomes. There is a well-defined interkinesis, but the formation of the daughter nuclei does not proceed very far. The homoeotypic division is passed through swiftly. Walls are formed around the spores in the typical manner. These walls later thicken and become sculptured and soon the anthers open and the pollen is discharged. The pollen nucleus divides sometime previous to the rupture of the anther.

Bremer reports incomplete reduction to be the cause of pollen sterility in some sugar cane. He found that many chromosomes failed to pair and that therefore pollen grains with different chromosome number could be found. These irregularities in chromosome number may cause the death of the gametophyte or impair the fertility of the male nuclei. But since in sugar beets no such irregularities exist, it cannot be considered a factor.

A differential rate in the growth of the pollen tube of foreign and own



pollen would also hardly be a factor since the pollen tube of the pollen of the sugar beet grows remarkably rapidly and has to traverse only a very short stylar canal. The ovule is commonly so oriented that its tip with the micropyle all but touches the roof of the ovary cavity and only a short distance away from the stylar opening. Sometimes, however, the tip of the ovule may have a more eccentric position and may even be turned slightly downward, making it more difficult for the pollen tube to reach it and thereby giving a more rapidly growing pollen tube the advantage.

Partial and entire degeneration of anthers and sometimes of the entire flower was often observed by the writer in material which harbored in the inflorescence axis or its lateral branches the encased larvae of insects. Due to the pathological changes in the tissues coincident with the presence of those insects, the food supply of the flower was cut off or at least impaired.

While self-fertilization in beets may often be frustrated because of the more or less protandrous condition of the flower, fertilization between neighboring flowers of the same plant undoubtedly does occur and as far as cytological evidence goes, such a union should not be disharmonic. But the fact that seed production in selfed individuals is greatly reduced shows a certain degree of incompatibility which, in all probability of a physiological nature, is too subtle to yield to a convenient analysis. Both Nillson and Dudok van Heel find this incompatibility more pronounced in some races than in others and that there appear to exist self-sterile as well as self-fertile lines.

It appears that substantial advances in beet improvement will only be made when we will use the individual flower as a unit in order to obtain genetically purer lines. That this is possible is shown by the fact that seed can be obtained from selfed flowers especially if one works with races which show the least incompatibility.



# MANIFESTATIONS OF IMPOTENCE IN A PLANT PROPAGATED BY SEED

D. F. JONES

*Connecticut Agricultural Experiment Station*

(WITH PLATES 22-23)

Inability to reproduce, due to a derangement of the floral organs or to a failure to produce viable germ cells, is a common occurrence in maize. Many of these manifestations of sterility, of the type classified as impotent by Stout (1916), are transmitted from generation to generation as segregates from normal plants or from individuals which are deficient in only one or the other of the sexual parts.

The nature and inheritance of these sterility factors have been described from time to time. Some of them destroy the male flowers, others the female, causing the plants to function as pistillate or as staminate individuals.

One factor is known which sterilizes both types of floral organs. This was first illustrated by East and Hayes (1912) from material furnished by R. A. Emerson. This factor has been called *naked* and from seed supplied by Dr. Emerson plants have been grown which are completely sterile. Usually the staminate flowers, as shown in FIG. 1, produce a few spikelets and develop a small quantity of good pollen. A few functional ovules are also produced on some of the lateral inflorescences and viable seeds are formed which when self-pollinated reproduce the sterile condition on all plants.

This transmissible form of sterility can be recombined with other characters and in FIG. 2 there is shown a *dwarf* form equipped with a naked tassel and ear. Generally the plants with the recessive *naked* factor in the homozygous condition are full sized and well developed in all other respects. In one  $F_2$  progeny, however, which resulted from a cross with a dwarf form known as *nana*, there appeared a number of plants similar to those shown in FIG. 3. These curious individuals were perfectly normal up to the fourth or fifth node. They had well-developed root systems, the stalks were as large in diameter as the normal plants in the same progeny which grew seven to eight feet tall and what few leaves that did develop on the plants were full-sized. At the fourth or fifth node development ceased as shown in the plant with the leaves removed. A small structure is shown at the tip of the plant and the tissue of which this was composed was similar to the sterile ears on the usual type of *naked* plants. Here is a case in which a factor, which usually has no effect until the flowers are formed, begins to operate at an earlier stage and stops normal development abruptly. Full-flowered dwarfs as well as naked dwarfs were obtained in this same progeny so that the unusual appearance of the naked factor in this one progeny does not seem to have any relation with the *nana* factor.

Several genes are known which cause maize plants to become wholly pistillate either in structure or function. Emerson (1920) has described two types of plants having both terminal and lateral inflorescences producing only female flowers. Other inherited characters which affect maize plants in a



FIG. 2. A recombination of the factors for naked flowers and dwarf plants.

similar way have since been found. When crossed with normal plants, normal monoecious individuals are produced in the first generation and the pistillate types reappear in the second generation as simple Mendelian recessives.

Plants with only terminal pistillate inflorescences can be produced in maize by growing the plants in a shortened period of daylight as in the greenhouse during the winter. Similar structures are produced occasionally on the tillers of plants which are otherwise normal. The floral development of maize plants is influenced by both external and internal environmental differences as is nearly every other part of the plant. But in the usual summer growing conditions there are unisexual plants which result from certain transmissible factors in the germplasm and these are quite constant in their appearance.

A commonly inherited character in maize is a condition of pollen abortion. The anthers are shrunk and contain poorly developed or no pollen grains. L. A. Eyster (1921) has described and illustrated a condition of this kind. The sterile spikelets are not so well filled as normal spikelets and remain flattened against the rachis as shown in PLATE 22. The tassels have a characteristic appearance. This condition is called by him *male sterile* and behaves as a single factor recessive.



Sterile tassels of this kind have been noted in a number of different kinds of maize and many of them are genetically distinct. They differ somewhat in degree of sterility. The most extreme forms rarely protrude their anthers outside the glumes and then only after the glumes have dried and fallen apart. Some produce a small amount of shrunken pollen and in some cases seed may be formed from this. In most cultures the character is quite clear-cut and intermediate forms are not observed. All these plants showing pollen sterility are functional females. The pistillate flowers seem to be affected in no way and abundant seed is formed when pollen from other plants is available.

The directly opposite condition in which the staminate inflorescences are fully fertile and the pistillate inflorescences completely sterile is found in a type called *silkless* described previously (Jones, 1925). These plants produce lateral inflorescences that have a well-developed spike but the ovules are all aborted. No stigmatic threads are formed and no seeds are produced. Sterile ears of this kind are shown in PLATE 23. This character has been transmitted to several generations through the pollen and has its basis in a single recessive factor which shows linkage and crossing over with one of the plant color factors. It is apparently not at all affected by the usual en-



FIG. 1. A sterility factor in maize which affects both staminate and pistillate flowers.



FIG. 3, at left. Naked plants in which development has stopped abruptly at the fourth node.

FIG. 4, at right. A pistillate type of maize which produces only seeds in the tassel.

vironmental differences and no intermediate forms have been found either in the original material or in out-crosses.

Other families produce no lateral inflorescences on some of the plants, whereas the remaining plants under the same conditions have well-developed ears. Barren plants of this kind are frequently seen which are due to unfavorable growing conditions. In some cases they seem also to be due to genetic factors but it is not clear that these operate directly to suppress the lateral pistillate inflorescences or are merely growth factors which retard development and the reduction is exhibited in a failure to produce seed.

Silkless and earless plants are functionally staminate types. When crossed with pistillate plants, such as the male sterile or tassel seed types, normal monoecious individuals are formed. The sterility factors recombine independently and produce monoecious, pistillate, staminate and neuter plants. No single-sexed individuals have been found which, when crossed with the opposite sex, will give only two sex types as regularly occurs in dioecious plants and bisexual animals but the material now on hand suggests that such a condition may eventually be obtained and indicates that dioecism is here actually being brought about by a suppression of one or the other floral parts in different individuals.

## LITERATURE CITED

- East, E. M., and Hayes, H. K., 1916. Heterozygosis in evolution and in plant breeding. U. S. Dept. Agric., Bur. of Plant Industry Bull. 243.
- Emerson, R. A., 1920. Heritable characters of maize. II. Pistillate flowered maize plants. Jour. Heredity 11: 65-76.
- Eyster, L. A., 1921. Heritable characters of maize. VII. Male sterile. Jour. Heredity 12: 138-141.
- Jones, D. F., 1925. Heritable characters of maize. XXIII. Silkless. Jour. Heredity 16: 339-341.
- Stout, A. B., 1916. Self and cross-pollinations in *Cichorium Intybus* with reference to sterility. Mem. N. Y. Bot. Gar. 6: 333-454.

## EXPLANATION OF PLATES

## PLATE 22

Pollen abortion is shown in the staminate inflorescence on the right.

## PLATE 23

Upper photo: Normal and silkless pistillate inflorescences of maize.

Lower photo: Same with husks removed. The three inflorescences shown at the right have their ovules aborted and the plants which bear them are functionally staminate.

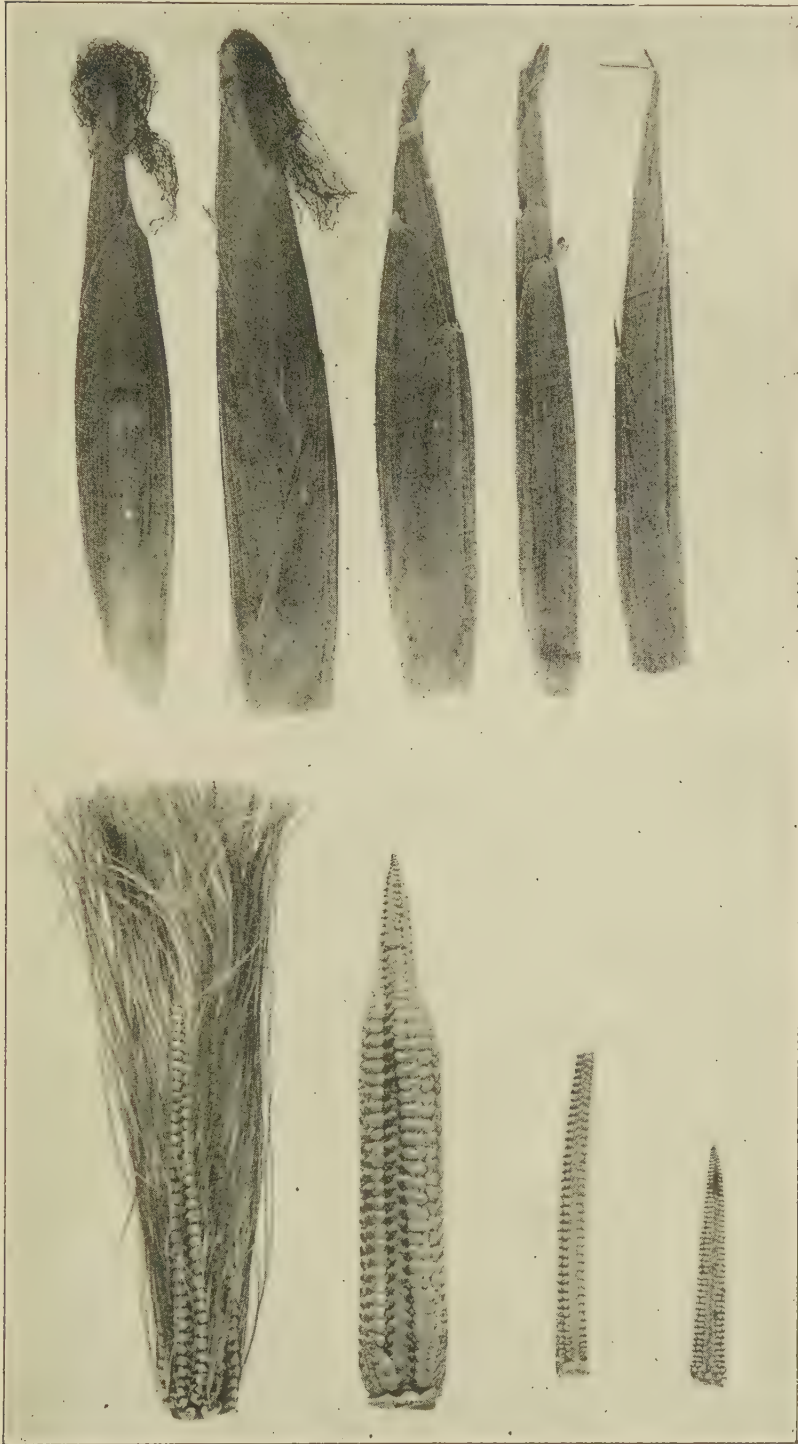






JONES: IMPOTENCE IN MAIZE





JONES: IMPOTENCE IN MAIZE





# STERILITY OF POLLEN IN DATURA

A. F. BLAKESLEE                      and              J. L. CARTLEDGE  
*Carnegie Institution of Washington      University of Pittsburgh*  
(WITH PLATE 24)

A number of factors have been found to be responsible for pollen sterility in *Datura*. The sterility may show itself in the formation of shrivelled grains, obviously incapable of germination. It may not appear, however, until tests are made of the ability of the pollen to germinate and to produce tubes capable of growing through the style. Furthermore, the generative nuclei must be able to fuse with the egg nuclei in order to produce viable zygotes. Physiological sterility, due to a gene, has been found in *Datura*, as well as that due to abnormal chromosomal constitution.

## POLLEN ABORTION IN DIFFERENT CHROMOSOMAL TYPES

Records have been given in an earlier publication (16) regarding the percentages of bad grains in different chromosomal types.

*Abortion in 1n, 2n, 3n, and 4n balanced types.* In PLATE 24 are photographs showing the abortion in balanced 1n, 2n, 3n, and 4n types. Summaries of the percentages of aborted grains are given in TABLE 1.

TABLE 1  
SUMMARY OF PERCENTAGES OF ABORTED POLLEN GRAINS. RECORDS OF 1923, 1924 AND 1925

	1n	2n	3n	4n	(2n + 1) TYPES	
					PRIMARIES	SECONDARIES
1923.....		1.6			3.0	7.7
1924.....		1.5			5.5	11.9
1925.....	88.0	1.2	43.6	5.2		

The normal diploid (2n) shows few bad grains, averaging only slightly more than one per cent. In the other even-balanced type—the tetraploid (4n)—the proportion is slightly higher—around five per cent—but the grains are distinctly larger as can be seen in the photographs. The pollen of the odd-balanced haploid (1n) is largely bad, with an average of around 88 per cent aborted grains. The full grains are all of the same size as those found in diploids. The pollen mother cells in haploids (4, 14) undergo a pseudo reduction into 6+6, 7+5, 8+4, resulting in most of the daughter cells being deficient for one or more chromosomes. A few normal grains are formed by non-reduction. Pollen of the odd-balanced triploids (3n) shows a higher proportion of bad grains, averaging around 44 per cent—and the full grains are of different sizes, from that seen in 4n to that seen in 2n plants. It is obvious that in haploids the aborted grains are caused by deficiencies in the normal chromosomal complement brought about by the process of pseudo

reduction. In diploids the percentage of bad grains may be caused by deficiencies brought about, in part at least, by detachment and by non-disjunction (3). The same causes may be effective in producing the higher percentages of bad grains in tetraploids in which non-disjunction has been shown to be common (5). In triploids, it has been shown that the chromosomes assort at random at reduction (3), producing pollen grains with deficiencies from the balanced  $2n$  number or additions to the balanced  $1n$  number. Detachment has been shown to be frequent in triploids (3). Chromosomal deficiencies, especially, or the unbalance due to extra chromosomes may be assumed to be the cause of bad grains in triploids.

### ABORTION IN UNBALANCED CHROMOSOMAL TYPES

Branches from otherwise diploid plants have occasionally been found which showed deficiency for a single chromosome (12). Such branches have given considerably more than the 50 per cent aborted grains expected, possibly due to the unbalance brought about in the environment of the surrounding somatic tissue.

TABLE 2

TYPES OF OFFSPRING THROWN BY  $2N$  AND PRIMARY AND SECONDARY ( $2N + 1$ ) PARENTS.  
FIGURES SHOW PERCENTAGES

	RELATED TYPES		UNRELATED	TOTAL OFFSPRING	TOTAL PARENTS
	PRIMARYS	SECONDARIES	( $2n+1$ ) TYPES		
Primarys.....	25.5	.14	.97	10,165	66
Secondarys.....	3.8	19.00	.99	8,754	64
$2n$ .....	....	....	.16	3,654	127

Primary ( $2n + 1$ ) types (6, 10) show an excess of bad grains over the amount shown by diploids (cf. TABLE 1). The excess is possibly due to the excess of non-disjunctional types thrown by the primarys. In a recent tabulation (TABLE 2) it was shown that in over 3,500 offspring from 127 diploid parents in Line 1A, a subline derived from a single haploid (13), there were produced 6 (or 0.16 per cent) non-disjunctional forms. From primary parents, which were in Line 1 or 1A, there were .97 per cent, and from secondarys, which were chiefly in Line 1 or 1A, there were .99 per cent. The influence of purely environmental factors, evident in the differences between the records of 1923 and 1924 shown in TABLE 1, has not been measured and the data on breeding behavior is too meager to warrant as yet a close statistical analysis. In working out formulae, therefore, we are striving rather to show a method of analysis than to arrive at a series of reliable constants. If  $d$  represent the percentage of grains resulting from detachment (3), and  $n$  the percentage of grains from non-disjunction, the formula for deficiencies, which we believe the chief cause of pollen abortion (P.A.) may be written:  $P.A. = d + n$ . The detachment ( $d$ ) affects, let us guess, around 0.5 per cent of the grains. Possibly only the  $n - 1$  grains, and not the minute microcytes

produced by detachment, would be recognized as aborted grains in pollen counts. In judging the amount of non-disjunction from the percentages of spontaneous occurrence of non-disjunctive types in the offspring, we must take account of the fact that not all of the  $(2n + 1)$  zygotes reach a recordable age. Roughly, about 25 per cent of the progeny of a primary are like the parent type (TABLE 2), instead of the 50 per cent theoretically possible. Hence, there are about three times as many  $(2n + 1)$  zygotes [or  $(n + 1)$  egg cells] as appear in the offspring. The .16 per cent of non-disjunctive types from  $2n$  parents shown in TABLE 2 must therefore be multiplied by 3 to get the actual percentage of non-disjunction. The formula then becomes for  $2n$ ,  $P.A. = 0.5 + .48 = .98$ , a figure somewhat less than the percentage of pollen abortion found for diploids in 1925.

Multiplying by 3 the .97 per cent new non-disjunctive mutants thrown by primaries, we have the formula for Primaries:  $P.A. = 0.5 + 2.91 = 3.41$ , a figure intermediate between the percentages of aborted grains found in primaries in 1923 and in 1924. The reason why secondaries throw a higher proportion of aborted grains than do their primaries is probably connected with the manner in which primaries are produced by secondaries.

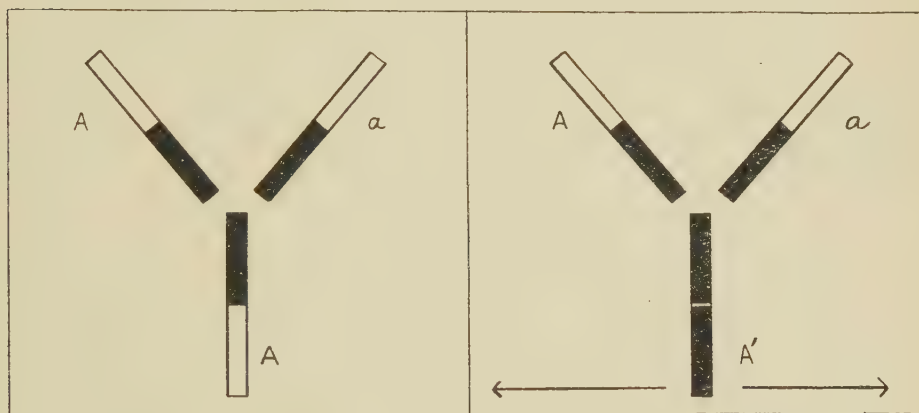


FIG. 1. Diagram of a trisome of a primary ( $2n + 1$ ) type at left, and of one of its secondary ( $2n + 2$ ) types at right. In the primary the chromosomes in the trisome contain each a "shaded" and a "white" half and assort at random. The pollen grains and egg cells should be  $2A + a + A_2 + 2Aa$ , all of which may be viable. In the secondary, the extra chromosome ( $A'$ ) contains two shaded halves but lacks a white half. If  $A$  and  $a$  go to opposite poles, the pollen grains and eggs should be  $A + a + AA' + aA'$ , all of which may be viable. If, however,  $A$  and  $a$  go to the same pole, the pollen grains and eggs should be  $A' + Aa$ .  $A'$ , since it lacks the white half of the chromosome, would probably die and be represented by an aborted grain or ovule.  $Aa$  would form an egg cell which, when fertilized, could give rise to the primary. The 3 chromosomes in the trisome of a secondary sometimes form a trivalent and sometimes a bivalent + a univalent, the bivalent apparently containing the two normal chromosomes and the univalent the double-half chromosome (6). Disjunction of the bivalent and assortment of the univalent should give normal and secondary gametes, while disjunction of a trivalent, if at random, should give normals and 1 primary to 2 secondary gametes. The relative proportions, therefore, of trivalents to bivalents + univalents might account for the ratio of primaries to secondaries in the offspring of secondaries.

In FIG. 1 at left is a diagram showing the three homologous chromosomes of a primary. The breeding evidence is good that in primaries the assortment of chromosomes is at random (15). In secondaries, in which the extra member of the set is a double-half chromosome, apparently the normal chromosomes usually go to opposite poles at reduction (FIG. 1 at right). Only when the two normal members go to the same pole should there be possible a  $(n + 1)$  primary pollen grain. The corresponding grain with the double-half chromosome, since it lacks the opposite half, would probably abort. The percentage of primaries thrown by secondaries (about 3.9 per cent) multiplied by 3 should therefore give a rough index of the proportion of aborted grains due to this process of disjunction. The formula for secondaries, therefore, should be:  $P.A. = 0.5 + (.99 \times 3) + (3.9 \times 3) = 14.9$ , a figure higher than the average percentage of abortive secondaries shown in TABLE 1. We have not considered the possibly greater mortality of pollen grains with a single extra chromosome, and in certain cases this should be taken into account. The previous discussion, however, shows that deficiencies, brought about chiefly by some abnormal form of disjunction, will largely account for the percentages of pollen abortion observed.

Since extra chromosomes are transmitted chiefly through the female and since chromosomal irregularities apparently may occur in somatic tissue, the breeding data are not directly applicable to pollen abortion. The differences in non-disjunction between diploids and  $(2n + 1)$  types, concluded from breeding data, are not confirmed by direct observation of the chromosomes (6). The figures, therefore, obtained from our formulae are to be considered only as provisional approximations pending more reliable data.

#### ABORTION IN HYBRIDS

Pollen abortion has been found in hybrids between species and by some authors is considered a proof of the hybrid origin of species which show a high proportion of pollen abortion. The classical case is that of hybrids between certain species of *Stizolobium* in which Belling (1) established the fact that half of the pollen grains, as well as half of the ovules are aborted. From his cytological study of a  $(2n + 1)$  type in *Datura* which occurred only in hybrids between "A" and "B" whites, it was suggested (7) that the peculiar breeding behavior of the "B" whites (11) might be explained by assuming segmental interchange between non-homologous chromosomes in the origin of "B" whites. This hypothesis was applied to the *Stizolobium* hybrids (2). The hypothesis would anticipate 50 per cent abortion in  $F_1$ 's between "A" and "B" whites and such abortion of both pollen grains and ovules was found in  $F_1$ 's between "B" whites and Line 26, which is an "A" white. Further tests, however, have shown that Line 26 will produce  $F_1$ 's with 50 per cent pollen abortion, when crossed with most of our lines—"A's" as well as "B's." Lines 1 and 26 have been used as testers. The majority of our lines, when crossed with Line 1, produces  $F_1$ 's with good pollen, but, when crossed with Line 26, produces  $F_1$ 's with 50 per cent abortion. Line 26 and these other "bad pollen



inducers" when crossed together produce  $F_1$ 's with good pollen. An attempt was made to locate the cause of the pollen abortion in specific chromosomes by backcrossing, to Line 26, the various primary ( $2n + 1$ ) types rendered heterozygous for Lines 1 and 26. All of the types threw 1:1 ratios of plants with good pollen to those with 50 per cent abortion, except Echinus and Microcarpic, which threw more than twice as many plants with 50 per cent abortion. It appears from these trisomic ratios (15) that the Echinus and the Microcarpic chromosomes are together, responsible for the 50 per cent abortion and the hypothesis of segmental interchange between non-homologous chromosomes would seem applicable in explaining the phenomenon. Such an hypothesis would lead one to expect to be able to find, in  $F_1$ 's, configurations with four chromosomes attached. A considerable number of slides have been obtained by Miss Rachel Haynes and Miss Louise H. Buck from buds of  $F_1$ 's between Line 1 and these bad pollen inducers. Although configurations were numerous, none were found showing the attachments expected. In  $F_1$ 's between Line 1 and "B" whites, however, such attachments were common, resulting occasionally in closed rings of four chromosomes of two different sizes. The problem is: why  $F_1$ 's between Line 1 and "B" whites do not show 50 per cent abortion and why the  $F_1$ 's between Line 1 and the Line 26 types do not show the configurations expected. If chromosomes which contain homologous parts tend to go to opposite poles at disjunction, one might expect the two abnormal chromosomes derived from the "B" whites to go to one pole and the corresponding normal chromosomes from Line 1 to go to the other, with no production of aborted pollen grains which a random assortment of these four chromosomes might be expected to bring about. If in the origin of each of the two abnormal chromosomes in Line 26, the outer end of one segment had become attached to the inner end of the other, closed rings of four chromosomes would not be expected. Further study, however, will be necessary to determine the actual chromosomal arrangement in "B" whites and in bad pollen inducers.

It is probable that several different chromosomes are responsible for the pollen abortion in  $F_1$ 's from *inter se* crosses of the lines in our collection. Thus a line from Angol, Chile, gives about 25 per cent abortion in  $F_1$ 's with Line 1. In this case the trisomic ratios seem to indicate that both the Cocklebur and the Globe chromosomes are together responsible for the abortion. Linkage seems to exist between the factor for pale stems located in the Globe chromosome and a chromosomal portion which shares the responsibility for the abortion.

A third type of bad pollen inducers is represented by our Line 27, which came from Mexico, but the chromosomes responsible for the pollen abortion have not yet been determined.

In the bad pollen inducers and in the "B" whites we have evidence of a not infrequent rearrangement of chromosomal material in the evolution of biotypes in nature.

### ENVIRONMENTAL FACTORS

Environmental factors may have a marked influence upon the production of aborted grains. It has already been shown (8) that the Quercina disease may prevent the formation of good pollen without having marked effect upon the production of ovules. The most striking environmental influence causing pollen abortion is cold (9). TABLE 3 is taken from a series of cold experiments started in 1921 and shows the proportion of bad grains after exposing plants to temperatures of an unheated greenhouse (50°-60°F.) TABLE 4 shows the measurements of pollen from several of these flowers. For reference, measurements of pollen grains made at the same time from flowers of the balanced types are also included and arrows below indicate the range for pollen from 2n, 4n flowers and for the pollen that would be expected from 8n plants, which have not yet been discovered. It will be seen that the effect of the cold has caused a large amount of abortion, probably through disturb-

TABLE 3

DATURA, LINE 1, COLD EXPERIMENT 5. BUDS EXPOSED TO COLD FROM 11/22/22 TO 11/29/22

FLOWERS OPENING	DISTRIBUTION OF FLOWERS ACCORDING TO PERCENTAGES OF BAD GRAINS IN POLLEN					
	0-5	6-10	11-30	31-50	51-70	71-100
12/7/22.....	2	..	..	..	..	1
12/8/22.....	..	..	..	..	..	2
12/9/22.....	..	..	4	2	9	9
12/10/22.....	14	1	3	1	1	
Untreated Balanced Types	2n. 4n			3n		1n

TABLE 4

DATURA LINE 1 MEASUREMENTS OF POLLEN GRAINS OF SELECTED FLOWERS FROM TABLE 3, SHOWING INFLUENCE OF COLD TREATMENT UPON SIZE OF GRAINS. ABOVE, FOR REFERENCE, ARE GIVEN MEASUREMENTS, MADE AT THE SAME TIME, OF GRAINS IN INDIVIDUAL FLOWERS OF RESPECTIVELY 1n, 2n, 3n AND 4n PLANTS. BELOW, ARROWS INDICATE THE SIZE RANGE OF GRAINS IN 2n, 4n FLOWERS AND THE RANGE TO BE EXPECTED IN 8n FLOWERS WHICH, HOWEVER, HAVE NOT AS YET BEEN DISCOVERED. A DIVISION IN THE SCALE USED EQUALS 3.3μ.

SCALE	13	14	15	16	17	18	19	20	21	22	23	24	25	26	27	% BAD GRAINS
1n	...	3	3	17	44	27	6	...	...	...	...	...	...	...	...	98.66
2n	...	8	55	34	3	...	...	...	...	...	...	...	...	...	...	0.20
3n	1	4	13	30	22	15	9	3	1	2	...	...	...	...	...	32.11
4n	...	...	...	...	...	2	20	24	40	14	1	1	...	...	...	2.25
Cold	...	...	...	...	...	...	...	...	...	...	...	...	...	...	...	...
a	...	...	37	59	3	...	...	...	...	1	...	...	...	...	...	2.67
b	...	1	5	53	33	5	...	...	1	2	...	...	...	...	...	17.23
c	...	...	29	48	19	...	...	2	...	...	...	...	...	...	...	40.06
d	...	5	16	31	10	3	7	8	13	4	2	1	...	...	...	60.05
e	...	...	...	...	8	6	8	13	23	29	13	1	...	...	...	65.17
f	...	...	7	9	18	8	6	8	13	11	9	9	2	...	...	84.20
g	...	...	...	1	...	1	1	6	8	19	20	19	20	4	1	84.47
h	3	12	30	36	15	1	1	...	...	1	...	...	...	...	...	87.86
i	3	7	20	16	8	4	8	14	10	10	...	...	...	...	...	95.45



ing the mechanism of reduction and bringing about deficiencies. In PLATE 24 are shown photographs of typical fields of pollen from some of the flowers recorded in TABLE 4. It will be noted from the photographs, as well as from TABLE 4, that size of some of the grains has been increased in many of the flowers, probably by increase in chromosome number. In some cases the grains are as large as would be expected in octoploids ( $8n$ ).

TABLE 5

DATURA, LINE 1. COLD EXPERIMENT 3. FIVE PLANTS IN COLD HOUSE FROM EARLY BUD STAGE THROUGH RIPENING OF SEEDS (2/20/22 TO 6/6/22)

FLOWERS POLLINATED	POLLEN	POLLINATION	NUMBER OF FLOWERS	CAPSULES SET	OFFSPRING			
					NORMALS ( $2n$ )	( $2n + 1$ ) NUMBERS	( $2n + 1$ ) PER- CENTS	$4n$
3/27—4/11	Bad	Self	36	2	4	0	0	0
3/27—4/11	Bad	×Good Pollen	8	4	8	3	27.27	0
4/12—4/26	Bad	Self	50	42	3412	98	2.79	2
4/26—4/27	Good	Self	2	2	429	3	0.69	0

There is evidence that cold may increase the production of abnormal chromosomal types in the offspring as shown by TABLE 5. Miss Betty Watt has investigated the stages at which reduction takes place leading to the production of egg cells in comparison with the stage at which reduction takes place in pollen mother cells. Reduction in pollen mother cells occurs when the buds are about 7 mm. long, while the reduction in archesporial cells occurs much later when the bud is around 25 mm. long. Since the pollen grains with abnormal chromosome numbers do not ordinarily function, disturbance of the reduction divisions must be brought about in relatively large buds, considerably after the reduction in pollen mother cells, if it is to have any influence in inducing abnormal types in the offspring.

It is to be hoped that, with a cold chamber, in which the temperature can be more closely regulated than was the case in the sample experiments described, we may be able to induce the production of abnormal chromosomal types in the offspring by the application of cold at critical stages of development. Other experiments, as well as those summarized in TABLES 3, 4 and 5, leave no doubt that cold is an environmental factor capable of inducing pollen abortion and as well the production of large grains. It will not be possible in the present paper to discuss the results of investigators who have found cold and other factors of influence on pollen sterility in other species.

### SUMMARY

It has been shown that aborted grains in pollen of  $1n$ ,  $2n$ ,  $3n$  and  $4n$  plants is brought about probably chiefly through chromosomal deficiencies. Primary ( $2n + 1$ ) types have given more unrelated non-disjunctional forms in their offspring than have normal  $2n$  parents. The higher percentage of non-disjunction in primaries is believed responsible for the higher percentage of bad pollen in primaries in comparison with diploids. Secondary ( $2n +$

2/2) types are believed to produce a proportion of bad grains, corresponding to the number of primaries which they throw in their offspring.

Fifty per cent abortion of pollen grains and ovules are found in  $F_1$ 's between the majority of the lines studied and certain "bad pollen producers" which themselves have good pollen. The cause of the 50 per cent abortion has apparently been located in two chromosomes (Echinus and Microcarpic). Another type of bad pollen producers gives 25 per cent pollen abortion in  $F_1$ 's with Line 1, and in this case two other chromosomes seem responsible. Segmental interchange between non-homologous chromosomes, which was suggested as explanation of "B" whites, may be cause of bad pollen producers.

Low temperatures may cause abortion of pollen grains and an increase in size of grains, due presumably to abnormalities in chromosome number.

#### BIBLIOGRAPHY

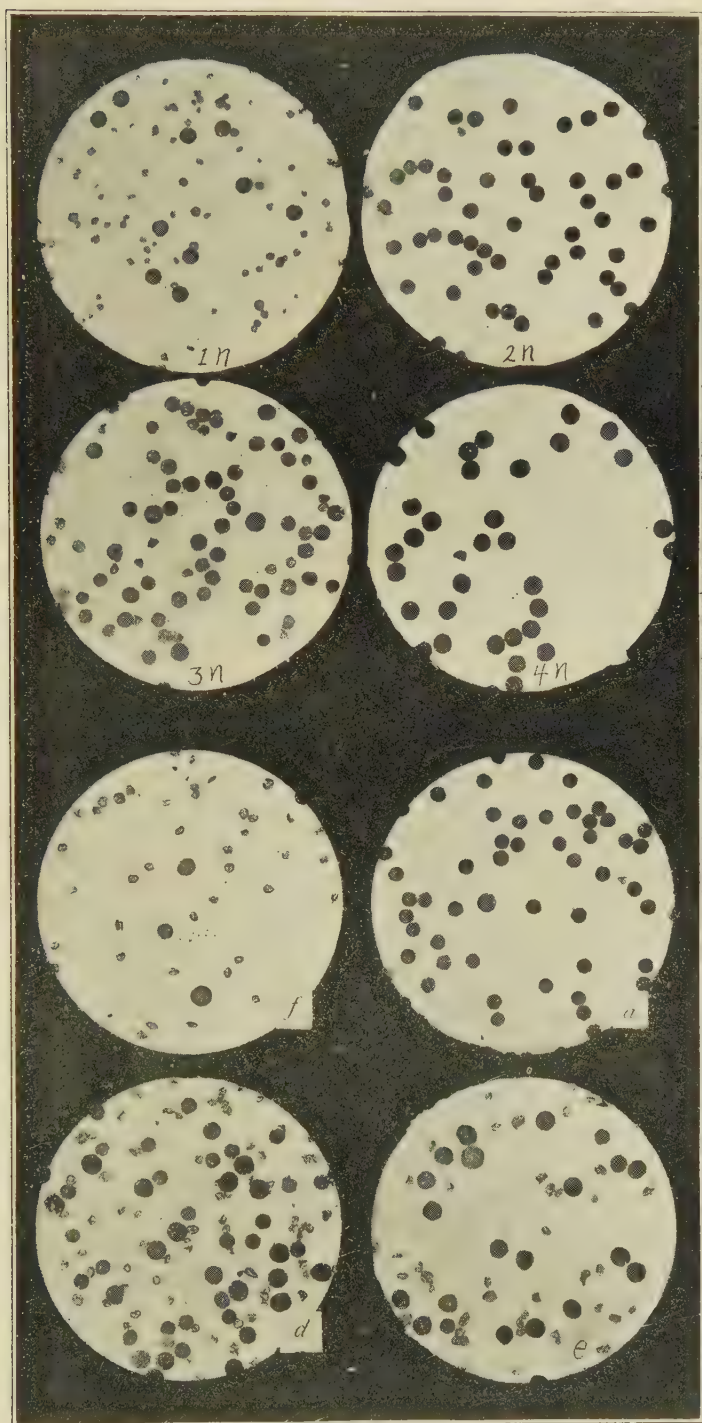
1. Belling, J., 1914. The mode of inheritance of semi-sterility in the offspring of hybrid plants. Zeitschr. f. ind. Abs. u. Vererb. **12**: 303-342.
2. ——— 1925. A unique result in certain species crosses. Zeitschr. f. ind. Abst. u. Vererb. **39**: 286-288.
3. Belling, J., and Blakeslee, A. F., 1922. The assortment of chromosomes in triploid daturas. Amer. Nat. **56**: 339-346.
4. ——— 1923. The reduction division in haploid, diploid, triploid and tetraploid daturas. Proc. Nat. Acad. Sci. **9**: 106-111.
5. ——— 1924. The distribution of chromosomes in tetraploid daturas. Amer. Nat. **58**: 60-70.
6. ——— 1924. The configurations and sizes of the chromosomes in the trivalents of 25-chromosome daturas. Proc. Nat. Acad. Sci. **10**: 116-120.
7. ——— 1926. On the attachment of non-homologous chromosomes at the reduction division in certain 25-chromosome daturas. Proc. Nat. Acad. Sci. **12**: 7-11.
8. Blakeslee, A. F., 1921. A graft-infectious disease of *Datura* resembling a vegetative mutation. Jour. Genetics **11**: 17-36.
9. ——— 1922. Quoted in Year Book of Carnegie Institution of Washington, No. 21, 1922, pp. 96-98.
10. ——— 1924. Distinction between primary and secondary chromosomal mutants in *Datura*. Proc. Nat. Acad. Sci. **10**: 109-116.
11. ——— 1924. Quoted in Year Book of Carnegie Institution of Washington, No. 23, 1924, pp. 24-27.
12. Blakeslee, A. F., and Belling, J., 1924. Chromosomal chimeras in the Jimson weed. Science, N. S. **60**: 19-20.
13. ——— 1924. Chromosomal mutations in the Jimson weed, *Datura Stramonium*. Jour. Heredity **15**: 194-206.
14. Blakeslee, A. F., Belling, J., Farnham, M. E., and Bergner, A. D., 1922. A haploid mutant in the Jimson weed, *Datura Stramonium*. Science, N. S. **55**: 646-647.
15. Blakeslee, A. F., and Farnham, M. E., 1923. Trisomic inheritance in the Poinsettia mutant of *Datura*. Amer. Nat. **57**: 481-495.
16. Blakeslee, A. F., and Cartledge, J. L., 1926. Pollen abortion in chromosomal types of daturas. Proc. Nat. Acad. Sci. **12**: 315-323.
17. Buchholz, J. T., and Blakeslee, A. F., 1922. Studies of the pollen tubes and abortive ovules of the Globe mutant of *Datura*. Science, N. S. **55**: 597-599.

#### EXPLANATION OF PLATE 24

The four above: Photomicrographs of pollen grains from a haploid (1n); from a diploid (2n); from a triploid (3n) and from a tetraploid (4n). Compare with the four below which show pollen from a diploid after buds had been exposed to cold.

The four below: Photomicrographs of pollen from flowers previously treated with cold. Letters indicate flowers listed in Table 4. Pollen of *a* is mostly normal, showing only large grains. Pollen of *c* shows a high percentage of aborted grains with most of the good grains larger than normal. Pollen of *d* shows a high percentage of aborted grains and a wide variation in size of good grains. Pollen of *f* shows mostly aborted grains and both large and normal-sized grains. Compare with the upper four which show pollen of 1n, 2n, 3n and 4n plants.





BLAKESLEE AND CARTLEDGE: Pollen in *Datura*



# THE HEREDITY OF SELF-STERILITY IN *VERONICA SYRIACA*

ERNST LEHMANN

*University of Tübingen, Germany*

(WITH PLATES 25-27)

We have had the good fortune to hear during the preceding sessions of this Conference of many important investigations on the phenomenon of self-sterility and we will have this afternoon the pleasure of learning about the more recent researches of the men to whom we owe many of the most interesting studies on this phenomenon.

The matter of which I am going to speak to you is the phenomenon of self-sterility or self-incompatibility. Will you kindly keep in mind that in this type of sterility the gametes while normal are unable to function in certain relations on account of physiological reasons which, however, are not entirely understood.

There are several points of view from which this phenomenon may be discussed and investigated. We may consider the practical significance of self-sterility, we may study it from the standpoint of physiology, and we may investigate its heredity.

In one of the earliest of the papers discussing self-sterility, Jost proposed an explanation from the doctrine of the development of individual stuff. It happens that in 1908, while I worked in Pfeffer's laboratory, during a discussion the paper of Jost was reviewed. Correns, who was then in Leipzig, at once advanced the idea of the hereditary transmission of self-sterility. Since that time many papers on the hereditary behavior of self-sterility have appeared. Partly they deal with the relation of self-sterility to self-fertility, partly they try to explain the heredity of self-and cross-sterility.

I shall not talk here about all the researches since that time. They will be reported much better today by some of the workers who have been concerned with these experiments. I shall limit my own paper entirely to the searches of myself and my pupil Filzer with *Veronica syriaca*. Also concerning these researches I must say that they are restricted to hereditary matters of self- and cross-sterility, and not extended to the relations between self-sterility and self-fertility. Nor do they deal with questions regarding the physiological processes involved.

But by keeping this in mind we shall see that the self-sterility of my experimental plant is very simple and we can rather easily comprehend its hereditary transmission.

The advantages of *V. syriaca* for the study of self-heredity are various: Firstly, I have found this plant always self-incompatible. I have never

found a single plant which was really self-compatible; nor has end-season fertility occurred. Thus many difficulties were avoided.

Secondly, the time required for the plant to reach maturity is very short. In about three to four months after sowing many hundred flowers may be crossed.

Thirdly, the crossing is very simple. The yellow pollen is always very easily seen on the whitish stigma.

I shall now proceed directly to consider our researches. I started by crossing two single plants from seeds obtained from the seed firm of Haage and Schmidt of Erfurt. From the  $F_1$  plants of this cross we pollinated mutually as many as possible. We observed very soon that not every combination was possible and that we got four groups of plants which we signified by the capitals A, B, C and D.

TABLE 1

RESULTS OBTAINED IN SELFING AND CROSSING SEVENTEEN PLANTS OF THE  $F_1$  GENERATION IN AS MANY COMBINATIONS AS COULD BE MADE

		A	B	C	D	A	B	A	B	D	D	B	B	A	C	A	A	A
		3	9	23	27	28	32	38	46	47	68	69	77	79	86	98	102	108
D	27	+	+	+	0	+	+	+	+	0	0	+	+	+	+	+	+	+
D	47	+	+	+	0	+	+	+	+	0	0	+	+	+	+	+	+	+
D	68	+	+	+	0	+		+	+	0	0	+	+	+	+	+	+	+
A	3	0	+	+	+	0	+	0	+	+	+	+	+	0	+	0	0	0
A	28	0	+		+	0	+	0	+	+	+	+	+	0	+	0	0	0
A	38	0	+	+	+	0	+	0	+	+	+	+	+	0	+	0	0	0
A	79	0	+	+	+	0	+	0	+	+	+	+	+	0	+	0	0	0
A	98	0	+	+	+	0	+	0	+		+	+	+	0	+	0	0	0
A	102						+	0	+	+	+	+	+	0	+	0	0	
A	108			+	+	0	+	0	+	+	+	+	+	0	+	0	0	0
C	23	+	+	0	+	+	+			+		+	+		0			
C	86	+	+	0	+	+	+	+	+	+	+	+	+	+	0	+	+	+
B	9	+	0		+	+	0	+	0	+	+	0	0	+	+	+	+	+
B	32	+	0	+	+	+	0	+	0	+	+	0	0	+	+	+	+	+
B	46	+	0	+	+	+	0	+	0	+	+	0	0	+	+	+	+	+
B	69	+	0	+	+	+	0	+	0	+	+	0	0	+	+	+	+	+
B	77	+	0	+	+	+	0	+	0	+	+	0	0	+	+	+	+	+

All the individuals of each group were incompatible with each other but compatible with all the plants of the three other groups. So we got the four intra-sterile but inter-fertile groups as shown in the following table.



TABLE 2

GROUP A	GROUP B	GROUP C	GROUP D
3	9	7	11
8	22	23	18
10	32	29	27
24	37	35	30
28	41	39	33
38	45	44	47
54	46	50	68
67	69	53	71
79	70	57	75
84	76	61	92
96	77	62	94
98	83	80	97
99	88	82	101
102	90	86	
108	91	106	
109	95	110	
Totals 16	16	16	13

In this chart sixteen different plants were in each of the groups A, B and C and thirteen were in group D. The four groups were most noticeably of nearly equal numbers.

We may show diagrammatically in PLATE 25 how these four groups arose. There are represented two flowers from the two plants which were crossed. You see represented the ovaries and the anthers and pollen, and you observe that the pollen tubes of a plant do not penetrate the pistils of that plant while the pollen tubes from the other plant do penetrate. Then the offspring of a cross fall into the four groups as discussed above.

In order to explain how these four groups arose we made various combination crosses between numbers of the various groups. That is, we crossed plants of group A with plants of group B,  $A \times B$ ,  $A \times C$ ,  $A \times D$ ,  $B \times C$ ,  $B \times D$  and  $C \times D$ . There are six such combinations possible.

Then we obtain very important and interesting results. In four cases the offspring fall into two intra-sterile but inter-fertile groups and in two cases there are four such groups. This condition may be summarized thus:

$$\begin{aligned}
 A \times B &= 2 \text{ groups} \\
 A \times C &= 2 \quad " \\
 A \times D &= 4 \quad " \\
 B \times C &= 4 \quad " \\
 B \times D &= 2 \quad " \\
 C \times D &= 2 \quad "
 \end{aligned}$$

As an example of these cross relations we give in the tables which follow the results of crosses between plants of the group  $A \times B$  (TABLE 3) and of the group  $A \times D$  (TABLE 4).

TABLE 3

THE RESULTS OF CROSS-POLLINATIONS AMONG PLANTS OF GROUP A  $\times$  B WHICH FELL INTO  
TWO INTRA-STERILE BUT INTER-FERTILE CLASSES

		II	II	II	I	II	I	I	I	I	II	II	I	II	II	I	I
		2	10	19	20	24	27	36	37	38	39	40	46	47	48	49	50
II	2	0	0	0	+	0	+	+	+	+	0	0	+	0	0	0	+
II	10	0	0	0	+	0	+	+	+	+	0	0	+	0	0	0	+
II	19	0	0	0	+	0	+	+	+	+	0	0	+	0	0	0	+
II	24	0	0	0	+	0	+	+	+	+	0	0	+	0	0	0	+
II	39	0	0	0	+	0	+	+	+	+	0	0	+	0	0	0	+
II	40	0	0	0	+	0	+	+	+	+	0	0	+	0	0	0	+
II	47	0	0	0	+	0	+	+	+	+	0	0	+	0	0	0	
II	48				+	0	+	+	+	+	0	0	+	0			
II	49	0	0	0		0		+	+	+	0	0	+	0	0		+
I	20	+	+	+	0	+	0	0		0	+	+	0	+	+	+	0
I	27	+	+	+	0	+	0	0	0	0	+	+	0	+	+	+	0
I	36	+	+	+	0	+	0	0	0	0	+	+	0	+	+	+	0
I	37	+	+	+	0	+	0	0	0	0	+	+	0	+	+	+	0
I	38	+	+	+	0	+	0	0	0	0	+	+	0	+	+	+	0
I	46	+	+	+	0	+	0	0	0	0	+	+	0	+	+	+	0
I	50	+	+	+	0	+	0	0	0	0	+	+	0	+		+	0

We will now ask how we can explain these quite uniform results of our crossings. We made the following hypothesis for this purpose.

1. Like Correns, we suppose that the rate of growth of the pollen tubes through the stigma is determined by Mendelian factors. So the cross-sterility and the self-fertility are gametically determined.

2. We suppose that in each plant two different factors, but only two factors, are transmitted through the gametes.

3. In this plant there are partly different, partly the same factors, but always these factors are lying at the same locus in the chromosome. They are like multiple allelomorphs which exclude each other. In each plant they are only in two homologous chromosomes.

4. The pollen-tube growth is inhibited always when the same factor is in the pollen as in the stigma or style.

With these suppositions in mind we can easily explain the whole hereditary transmission of self-sterility in *V. syriaca*.

We now return to our first crossing. We will explain the results with the use of the diagram of PLATE 26. You see on this plate the same flowers as before and from each flower the ovary and the anther. According to our hypothesis, we find in each of them two different factors which control the self-sterility; we represent the factors of one plant in red (*a*) and yellow (*b*),

and of the other one in blue (*c*) and white (*d*). Neither plant can be self-fertilized because its stigma has the two factors *a* and *b* or *c* and *d* of which the respective pollen carries at least one in each grain. But both plants have different sets of factors and, according to our hypothesis, the pollen of each can produce tubes that penetrate the stigmas of the other and lead to fertilization. In each cross they reach all the ovules and thus in both reciprocal crosses the same four combinations are realized. Both plants have two different factors and according to our hypothesis when the two plants are crossed all the pollen tubes can penetrate the stigmas. They reach all the ovules and thus give in both reciprocals the combinations *ac*, *ad*, *cb* and *db* which give the four different groups of plants. I have tried to show this by the color combinations: red and blue gives lilac (A of PLATE 25); white and red gives rose (B of PLATE 25); blue and yellow gives green (C of PLATE 25), and yellow and white gives paler yellow (D of PLATE 25). Naturally where the mating took place by chance we must get groups of about the same size which has already been shown.

But let us now see how our explanation will fit with the crossings of the members of the four groups which we made and from which we found that there came in some families two and in the other four intra-sterile groups.

TABLE 4

THE RESULTS OF CROSSES BETWEEN PLANTS OF THE GROUP  $A \times D$  SHOWING THE FOUR CLASSES OF PLANTS INTRA-STERILE BUT INTER-FERTILE

[illegible]

We will consider first the cross between the members of the groups A and B and refer to the left side of PLATE 27.

A plant of the A group has, according to our hypothesis, the gametic combination  $a$  and  $c$ , and B has  $a$  and  $d$ . Of the pollen of the individuals of the group B, the  $a$  pollen will not germinate on the stigma of A because the plants of that group have factor  $a$ . The pollen  $c$  will germinate and penetrate the style and reach the ovules in the style tissues because this factor is lacking in the style tissues. Two groups of progeny will result, one from the combination  $ad$  and one from  $dc$ . The reciprocal cross gives quite the same number of the inter-sterile groups, although the genotypical composition is different as can very well be proven by additional crosses but of which we will not speak now.

But now we must try to explain the cross between A and D which gives four groups. You see that here the plants have all the four factors different. A had  $ac$ , and D has  $bd$ . It follows that four groups will be obtained because none of the factors of the pollen are the same in any one of the pistil and ovules. The four combinations  $ad$ ,  $ac$ ,  $cd$  and  $cb$  are all possible as shown in the right side of PLATE 27.

Here we make a survey of all the six different possible combinations between the four groups, A, B, C and D we see easily, that always where the plants of two groups have one factor in common we get only two groups in the offspring but when all the four factors are different, then we get four groups as follows:

$$\begin{aligned} A (ac) \times B (ad) &= ad \text{ and } cd \\ A (ac) \times C (bc) &= ab \text{ and } bc \\ A (ac) \times D (bd) &= ab, ad, bc \text{ and } cd \\ B (ad) \times C (bc) &= ab, ac, bd \text{ and } cd \\ B (ad) \times D (bd) &= ab \text{ and } bd \\ C (bc) \times D (bd) &= bd \text{ and } cd \end{aligned}$$

I will only say that we also made many of the other possible cross-combinations as  $ab \times ab$ , or  $ab \times cb$  and that all the results accorded with our suppositions.

But there remains one point of view which we have not yet discussed.

We used in the results just reported only two plants as parents in the beginning of our crossings, with two different factors in each for self-sterility. But let us take now more plants of the seed of *P. syriaca* and let us make as many as possible cross-pollinations between them and see what will happen.

We find in most of the combinations cross-fertility. For example, we obtained in 800 combinations between 51 plants only 16 cases of cross-sterility. The question is now, how can we make this agree with our theoretical conclusions. For explanation we choose two other plants III and IV out of the original seed and crossed them at random. Again the  $F_1$  progeny usually fall into four groups of intra-sterile plants with rarely two such groups. Then if we cross the members of these groups of offspring from



III  $\times$  IV with the offspring of the cross I  $\times$  II we get in most cases cross-compatibility. This occurs when four factors in both plants are different; perhaps  $a, b, c$  and  $d, e, f, g$  and  $h$  as shown in 1 of TABLE 5. Also when two factors are the same in the plants crossed we still get cross-fertility as shown in 2 of TABLE 5. When one factor is different in both progenies to be crossed, out of sixteen combinations possible, we obtain cross-sterility in two cases (see 3 in TABLE 5).

TABLE 5  
DIAGRAMS SHOWING RESULTS WHEN PROGENIES TO BE CROSSED DIFFER IN THE NUMBER OF COMMON FACTORS

1															
		$ab$	•	$cd$						$ef$	•	$gh$			
A		B		C		D				$A^1$		$B^1$		$C^1$	$D^1$
$ac$		$ad$		$bc$		$bd$				$eg$		$eh$		$fg$	$fh$
								$ac$	$ad$	$bc$	$bd$				
								$eg$	$B+$	$B+$	$B+$	$B+$			
								$eh$	$B+$	$B+$	$B+$	$B+$			
								$fh$	$B+$	$B+$	$B+$	$B+$			
								$fh$	$B+$	$B+$	$B+$	$B+$			

2															
		$ab$	•	$cd$						$ef$	•	$cd$			
A		B		C		D				$A^1$		$B^1$		$C^1$	$D^1$
$ac$		$ad$		$bc$		$bd$				$ec$		$ed$		$fc$	$fd$
								$ac$	$ad$	$bc$	$bd$				
								$ec$	$B+$	$B+$	$B+$	$B+$			
								$ed$	$B+$	$B+$	$B+$	$B+$			
								$fc$	$B+$	$B+$	$B+$	$B+$			
								$fd$	$B+$	$B+$	$B+$	$B+$			

3															
		$ab$	•	$cd$						$ed$	•	$ab$			
A		B		C		D				$A^1$		$B^1$		$C^1$	$D^1$
$ac$		$ad$		$bc$		$bd$				$ad$		$bd$		$ae$	$be$
								$ad$	$ae$	$bd$	$be$				
								$ac$	$B+$	$B+$	$B+$	$B+$			
								$ad$	0	$B+$	$B+$	$B+$			
								$bc$	$B+$	$B+$	$B+$	$B+$			
								$bd$	$B+$	$B+$	0	$B+$			

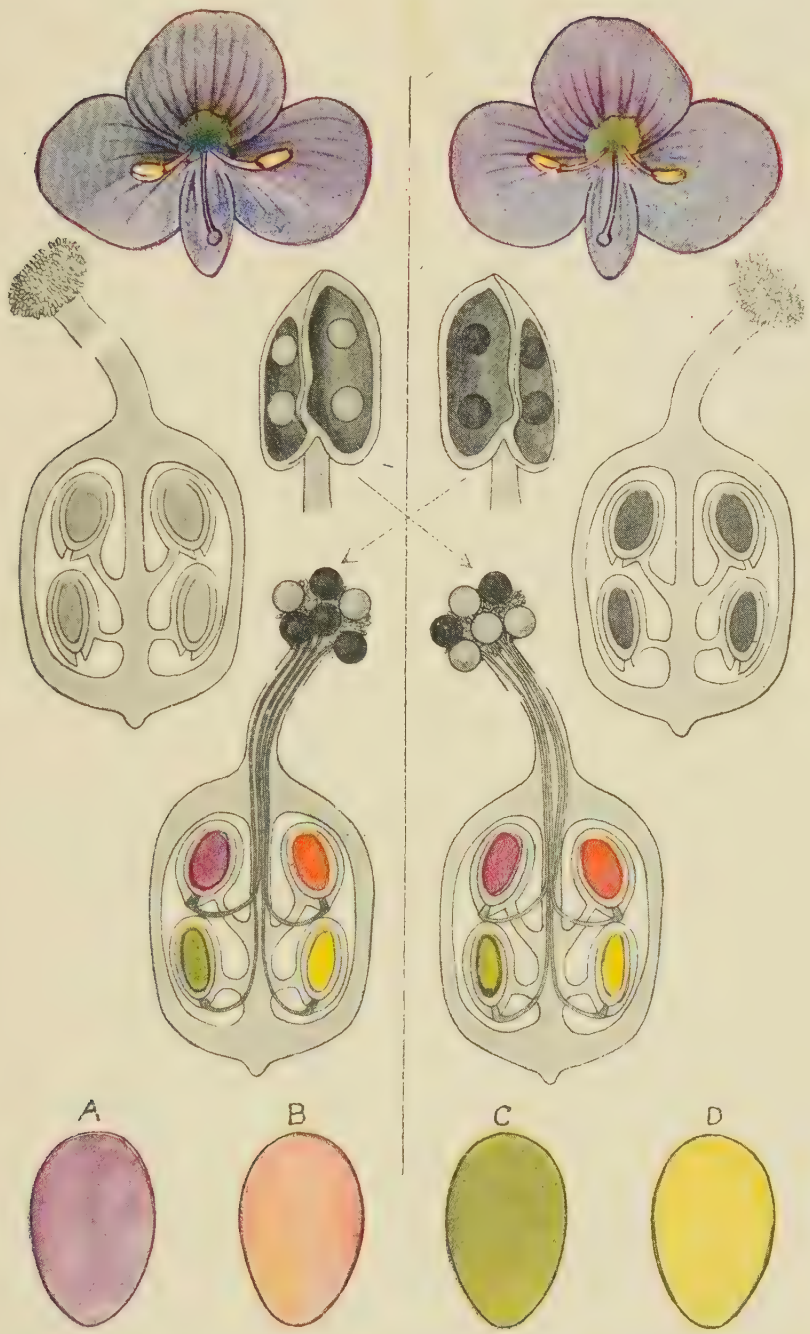
We could now find among our fifty-one plants which we used for our matings seven of such different factors behaving as multiple allelomorphs. Numerous crossings, many more than 50,000, proved always this hypothesis.

So we could prove our hypothesis and we could show that the self-sterility of *V. syriaca* is inherited through different multiple allelomorphs which inhibit or accelerate the rate of growth of the pollen tubes through the style.

Filzer was able to show from the results of Baur that in his researches of self-sterility *Antirrhinum* behaves in the same manner and I think we hear today that also in other genera the same hereditary transmission is observed.

#### EXPLANATION OF PLATES 25-27

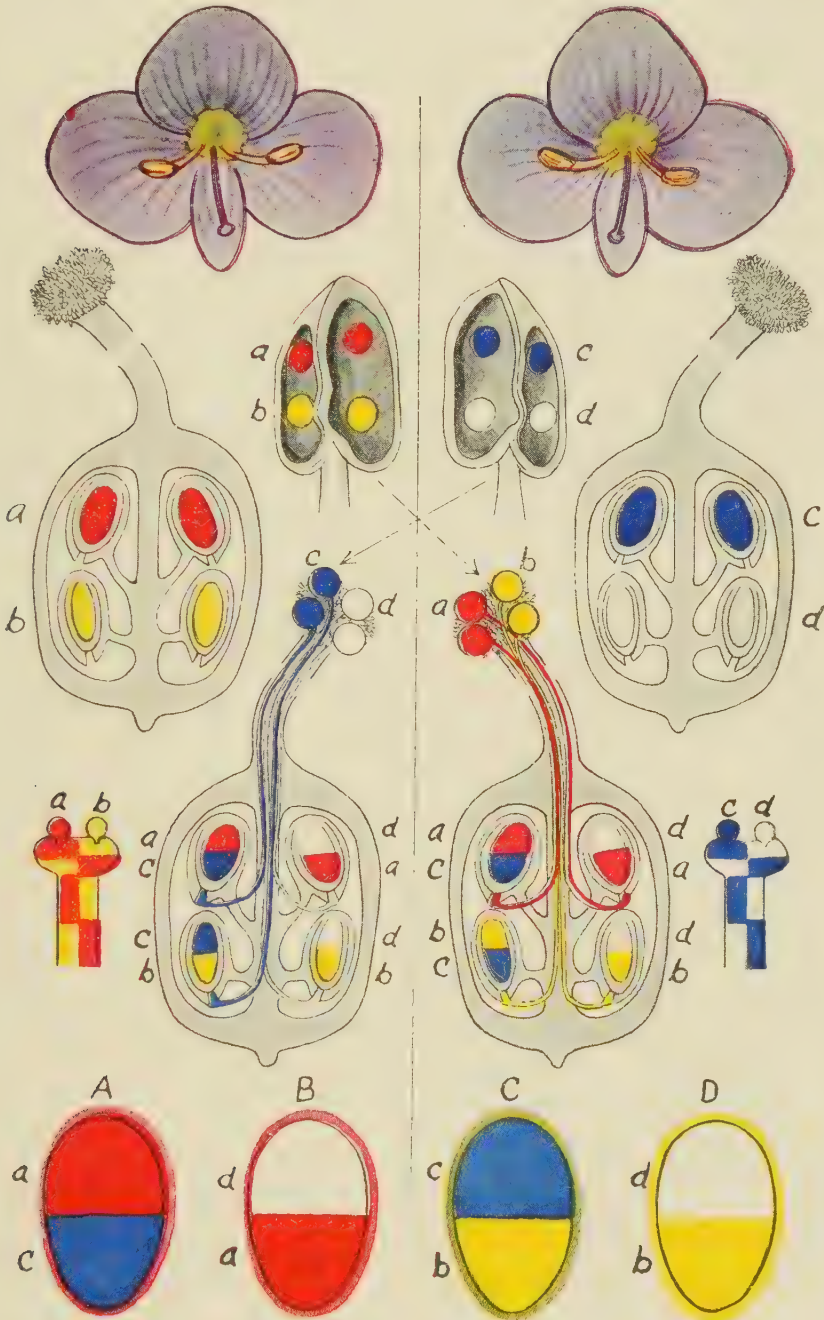
These three plates show diagrammatically how the self- and cross-fertilizations operate in respect to compatibility according to the theory of factorial control as expressed in this text.



LEHMANN: VERONICA SYRIACA



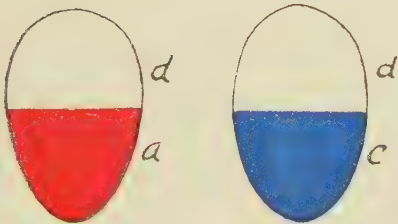
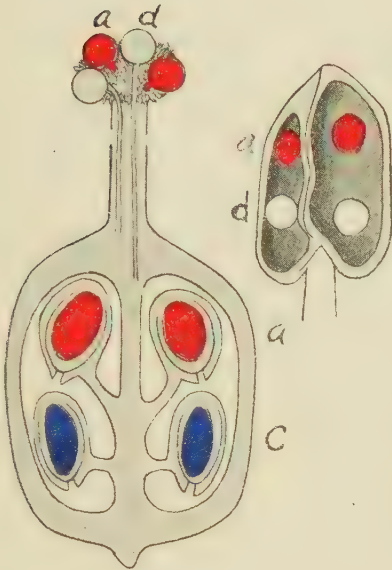




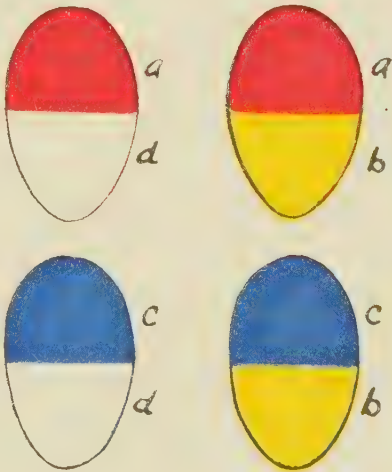
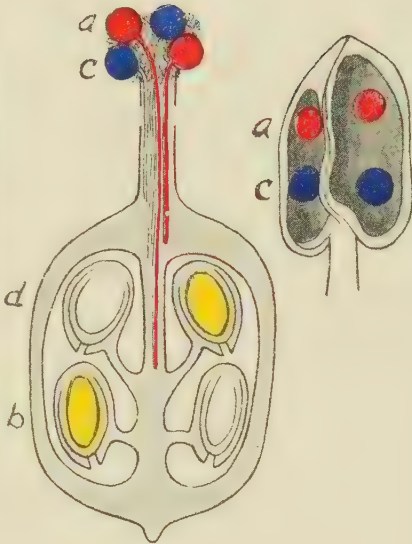
LEHMANN: VERONICA SYRIACA



A x B



A x D







## THE GENETICS AND PHYSIOLOGY OF SELF-STERILITY IN NICOTIANA

E. M. EAST      and      A. J. MANGELSDORF  
*Harvard University*      *Bussey Institution*

Although investigations have been under way on the problem of self-sterility at the genetics laboratory of the Bussey Institution of Harvard University for the past fifteen years, using *Nicotiana alata* and *N. Sandaræ* as material, it was only recently that sufficient genetic tests had been made to permit the formulation of a theory that interpreted satisfactorily the behavior of self-sterile plants in crosses where only self-sterile individuals were involved. This theory was published in the Proceedings of the National Academy of Sciences\* in February, 1925, but since it is not generally known, it may not be out of place to outline it here.

In endeavoring to analyze the mechanism which controls the behavior of these self-sterile plants, the chief obstacle was the difficulty of obtaining sufficient quantities of selfed seed, because only by the use of seedlings obtained by selfing can certain essential tests be made. When, a few years ago, it was found that nearly all of these particular self-sterile plants could be selfed by pollinating very young buds, therefore, it was felt that the strictly genetic problem of self-sterility stood some chance of being solved. It had previously been found that self-incompatibility and cross-incompatibility were due simply to a slow rate of pollen-tube growth and not to a true incompatibility between the gametes which would prevent zygote formation after the male nucleus had entered the micropyle of the embryo sac.

By taking advantage of this procedure, therefore, numerous families were produced among the so-called *Sandaræ* hybrids which consisted of two groups of progeny, all members of each group being sterile with each other but fertile with all members of the other group. A large series of crosses was made between the members of these various classes. The results obtained were similar. Let us take, for example, three classes which we may call A, B and C. Class A ♀ × Class B ♂ produced equal numbers of plants of Class B and Class C. The reciprocal cross, Class B ♀ × Class A ♂, yielded an equal number of plants of the two classes, A and C. Similarly, Class A ♀ × Class C ♂ gave an equal number of plants of Classes B and C. The reciprocal cross gave an equal number of plants of Classes A and B. A large number of such crosses, yielding without exception the same type of result, led us to the conclusion that the behavior of these three classes could be accounted for under the assumption that three members of one allelomorphic system,  $S_1$ ,  $S_2$  and  $S_3$ , here control the phenomenon of self-sterility. Class A is  $S_1S_3$ ,

---

\* East, E. M., and Mangelsdorf, A. J. A new interpretation of the hereditary behavior of self-sterile plants. Proc. Nat. Acad. Sci. 11: 166-171. 1925.

Class B is  $S_1S_2$  and Class C is  $S_2S_3$ . If, then, it is assumed that a plant affords stimulus only to pollen which bears sterility factors other than those found in its own tissues, all the observed facts are satisfied.

The ordinary type of plant found in self-sterile cultures is naturally heterozygous for the self-sterility factors carried; and very often when dealing with related plants, one sterility factor is common to both plants. A plant  $S_1S_2$  may be crossed with a plant  $S_1S_3$ . If, then, in the competition which ensues among the pollen grains of plant  $S_1S_3$  those bearing the factor  $S_3$  grow much faster than those bearing the factor  $S_1$ , equal classes of zygotes will be produced having the constitution  $S_1S_3$  and  $S_2S_3$ . This is, indeed, what is found. The reciprocal cross, however,  $S_1S_3 \times S_1S_2$ , results in equal-sized classes which prove to be  $S_1S_2$  and  $S_2S_3$ . In other words, when both plants are heterozygous and one sterility factor is common to both, the results of reciprocal crosses are unlike and the class of the mother is never obtained.

It may happen, however, that the self-sterility plants used in matings have no sterility factor in common; in such a case, the results of a reciprocal cross are identical. For example, when a plant  $S_1S_2$  is crossed with a plant  $S_3S_4$ , four equal-sized classes are produced which can be proved by proper testing to have the formulæ  $S_1S_3$ ,  $S_1S_4$ ,  $S_2S_3$  and  $S_2S_4$ . Each of these four classes is different from the class of either parent and is, therefore, fertile reciprocally with them.

A very interesting and critical test can be devised to prove or disprove this hypothesis. If a heterozygous plant—for example, a plant having the formula  $S_1S_2$ —is fertilized with its own pollen in the very young bud, there will be no competition between the pollen-tubes because the two types grow equally slowly; yet seed will be obtained, owing to the extraordinary length of time for pollen tube growth, and owing possibly also to the fact that pollen tubes grow more rapidly in young bud pollinations of this species. Ordinary Mendelian recombination should occur, therefore, yielding homozygous  $S_1S_1$  and  $S_2S_2$  plants, as well as the parental class  $S_1S_2$ . Plants of the two homozygous types should be fertile together, and should give the same type as that of the mother when crossed together. Each of these homozygous classes should be fertile as females with the heterozygous class  $S_1S_2$ , yielding a single class  $S_1S_2$ , but both homozygous classes should be sterile with the heterozygous class when used as males. The reason for this odd result is perfectly simple, according to the hypothesis. The homozygous  $S_1S_1$  plants will cross reciprocally with  $S_2S_2$  plants because the factors are unlike and every individual produced will have the formula  $S_1S_2$ . Naturally, however, neither the  $S_1S_1$  pollen nor the  $S_2S_2$  pollen will function on the heterozygous plants  $S_1S_2$  because in each case like factors are involved. On the other hand, in the reciprocal crosses  $S_1S_1 \times S_1S_2$ , or  $S_2S_2 \times S_1S_2$ , the entire resulting progeny will belong to the Class  $S_1S_2$  because in the first case only  $S_2$  pollen functions and in the second case only  $S_1$  pollen functions.

All of these tests have been made and have fulfilled expectation in a really remarkable way. Seven allelomorphs have been isolated and homozygous

"testers" produced which can be, and have been, used in further tests. That these factors are all members of one allelomorphous system—that is to say, that they are mutations at one locus—is shown first by the fact that when any two enter a given cross, only those two are recovered, and second by the fact that each is linked in inheritance with a certain flower-color factor and gives similar crossover values. Thus far no second locus, effective in self-sterility, has been found. We may, therefore, assume either that *like* factors produce substances that inhibit pollen-tube growth, or that *unlike* factors produce substances that accelerate pollen-tube growth. For various reasons the second assumption seems more probable; but speaking philosophically, if only one allelomorphous series of factors affecting pollen-tube growth is involved, it makes no difference which interpretation is adopted.

We have been greatly interested to find that the various data of Erwin Baur on self-sterile species of *Antirrhinum* yield to an analysis with the postulates given above. The recent work of Professor Ernst Lehmann and the work of Professor M. J. Sirks on *Veronica syriaca* and *Verbascum phoeniceum* respectively—reported in the proceedings of this conference—also fall into line. Doubtless cases of self-sterility will be found which do not yield so readily to analysis. Perhaps real differences of mechanism will be found; but since plants so different as *Nicotianas*, *Antirrhinums*, *Veronicas* and *Verbascums* show behavior which accords with the theory outlined above, we are inclined to feel that seemingly anomalous behavior in other genera is more likely to be due to additional variables being involved than to fundamental differences in the controlling mechanism.





## THE GENOTYPICAL PROBLEMS OF SELF AND CROSS-INCOMPATIBILITY

M. J. SIRKS

*Instituut voor Plantenveredeling, Wageningen, Holland*

The distinguished invitation, addressed to me by the organizing committee of this Conference, to take part in its program, was very welcome as it afforded me an opportunity to meet personally a great number of prominent scientists in the Eastern United States who are devoting their skill and activity to the solution of the interesting problems of genetics and of plant breeding. Thanks to the kindness of your committee I am now enabled to see their work and their researches as vivid things, while most of my fellow-scientists at the other side of the Ocean must be content with the printed results, which give only a superficial aspect of the work. During a number of centuries old Europe has abandoned many of its best individuals to what we call the New World; in Europe we consider it a duty of thankfulness to acknowledge the fact that the offspring of these emigrants have repaid this old debt copiously to those who stayed behind. European science is greatly indebted to American scientists.

Among the various problems afforded by the phenomena of unsuccessful pollinations in plants, there is one group that may be considered as a typical example of American-European collaboration as scientists in both parts of the world have contributed to its solution. It is that group of problems which are connected with the inheritance of self- and cross-incompatibility.

I do not intend to trouble you with a summary of all those manifold phenomena, which are in practice mixed under the name of sterility; two principles, however, may be separated as sharply as possible; that of true sterility and that of incompatibility.

Sterility is the phenomenon, to be found in cells and among groups of cells, that at a certain stage of their life the organization of these cells or cell groups is disturbed by internal causes, so that they abort or degenerate. This sterility may appear in gametes as well as in zygotes and in the organisms developing from the latter.

Incompatibility strikes gametes only which are wholly sound and functional, as for example pollen grains, while a disharmony between the organization of the tissue of the styles and their own inhibits the normal growth of the pollen tubes. If it happens, however, that such pollen-grains meet with a stylar tissue in which their growth is not affected, their development will be a normal one and their tubes will reach the aim of their existence: the fertilization of the ovule.

In earlier times this incompatibility was not kept separate from the

true sterility and it was considered to affect only pollen originating from the same plant, to which the stylar tissue belonged; so we are accustomed to speak of self-sterility while self-incompatibility is meant; since, however, we have observed that this incompatibility may also inhibit the growth of pollen from other individuals of the same species we may join this as cross-incompatibility as another type of the same physiological process.

The problems afforded to the geneticist by this self- and cross-incompatibility are many and various, but in substance they can be reduced to three main points. First, we must find an answer to the question, if this incompatibility is caused by the presence of genotypical factors, and if so will then these factors behave themselves according to Mendelian rules? Secondly, are these genotypical factors fixed and unalterable, are they units which are inherited from parents to children, and will they remain always the same during the successive generations? Third problem: what are the relations between self-incompatible and self-compatible plants and how may we consider these relations under the light of the other problems?

The researches that may serve for a reply to these problems are not yet fifteen years old. Before 1912 the problem of self-incompatibility was considered to be of flower-biological nature only, and Focke, Hildebrand, Jost and others have contributed many valuable studies to this side of the matter.

Another way, however, has been found by Correns (1912, 1913), who applied the methods of Mendelian genetics to the problem of self-incompatibility, a study furnishing marked and important data. A number of other scientists have followed his example, while they worked along the same lines with other species of plants and obtained results, partly corresponding to those of Correns, partly, however, in sharp contradiction to them: Correns, as the first with *Cardamine pratensis* (1912, 1913), and later with *Linaria vulgaris* (1916), the secretary of our conference, Dr. Stout with *Cichorium Intybus* (1916, 1917, 1918, 1920, 1923) and afterwards with other species as *Eschscholtzia californica* (1920), East and his collaborators, Park and Anderson (1915, 1917, 1918, 1919, 1923, 1924, 1925), with individuals of *Nicotiana glauca* and with hybrids between *N. glauca*, *N. glauca* and *N. glauca*, Heribert Nilsson (1916) with rye, Lehmann (1919, 1922) with *Veronica syriaca*, Baur (1919) with *Antirrhinum hispanicum*, Terao (1923) with *Petunia violacea*, Wada (1923) with the sweet potato, *Ipomoea Batatas*, and I myself with *Verbascum phoeniceum* (1917, 1926).

The data produced by these experiments were not always wholly reliable, especially in the earliest publications. The technical difficulties which on superficial consideration may seem to be slight, are indeed in some cases rather important. The success of a pollination does not only depend on the fact that compatible pollen is brought upon the pistil; temperature and humidity of the environment, the age of the pistil and that of the pollen, and the age of the plant itself are factors which may make it difficult to obtain a definite answer on the question if the pollen used may be compatible or not.

When the method of isolation is unfavorable, so that the atmosphere in

which the flowers are living becomes too warm and too humid, then a pollination will not result in a fertilization of the ovules, though the pollen as such may be able to function in a normal way. When, however, the method of isolation is not absolutely safe, which may be so in using small bags made of parchment paper or muslin, then the possibility is produced of a fertilization, caused by foreign pollen without our influence. When the bud is too young and the style of the flower has not yet reached its maximum of development, the fertilization may be obtained by pollen which is wholly incompatible in the styles of out-grown flowers. In some cases the age of the plant itself is of a rather great importance: East found in his plants of *Nicotiana* in some cases a strong end-season self-compatibility, in other plants the same phenomenon in a less striking form. Stout observed the same process in *Lythrum Salicaria*, while in *Brassica pekinensis* he found just the inverse, a mid-season self-compatibility; according to Baur's observations individuals of *Antirrhinum latifolium* in the first year showed a marked self-incompatibility, with a very slight end-season self-compatibility, while these same individuals became rather self-compatible in the second year of their existence. In *Verbascum phoeniceum* the self-incompatibility seems to be very strong; I myself never observed any trace of self-compatibility neither in any part of the season, nor in later years. Plants of eight years old are now still fully self-incompatible, just as they were in their first year. The age of the flower, however, in *Verbascum* is of a certain importance; young flowers immediately after their opening may be fertilized by pollen from older ones of the same plant; on the second day of life of these flowers, such a pollination remains without any result.

If, however, all possible care is taken to avoid any mistake, then it will be possible to get a view into the inheritance of the phenomena of self- and cross-incompatibility along the lines indicated by Correns; a theoretical explanation may be given that corresponds rather well with the data observed. First, therefore, I will try to summarize in a few words the work done by some of the workers mentioned above.

Correns worked with the meadow-cress, *Cardamine pratensis* and crossed two plants B and G, from a wholly distant origin; all the individuals of the  $F_1$  generation (2 reciprocal groups of 30 plants each) were pollenized with pollen from both parents and with that of twelve of their sibs. According to their behavior toward both parents, the  $F_1$  plants could be divided into four groups: a number of plants were capable of being fertilized by pollen of both parents; a second group by pollen of parent B only; the third group brought seed only when pollenized with pollen of the other parent G and the fourth group was incompatible with both parents. This same division into groups seemed to appear again in the results of the pollinations with pollen from the sibs. But the division may be considered to be of a rather rough nature; in the table, which is constructed by joining two of Correns' tables, a number of irregularities may be observed. First, the answer to the question if a certain pollination was successful, is not always without doubt, for

Correns qualifies the results of a pollination by very different gradations: very good, good, rather good, sparingly, bad, very bad, hardly anything, one seed only, nothing. In the table given here the terms very good till sparingly, inclusive, are indicated by a +, bad till nothing by a minus. Furthermore a number of pollinations are registered, which should be classified according to some results in the minus-class, according to others, however, in the plus-class. One of the individuals (2d) has even produced a favorable result after self-pollination and seemed thus to be self-compatible.

TABLE 1  
CORRENS ON *Cardamine pratensis*, 1912  
JOINED TABLES 1 AND 8  
MALES

	B	G	bg	lx	11	1ae	2d	2e	Bg	BG	2t	
	+	+	+	+	+	+	+	+	+	+	+	
1c	4	5	---	3	3	2	2	2	2	1	3	bg
1o	6	5	2	2	1	1	1	2	2	1	3	
1p	3	1	4	3	6	2	1	2	3	5	3	
1r	7	2	3	2	2	1	2	2	3	3	3	
1s	8	4	3	3	3	1	1	1	3	4	3	
1v	6	4	3	3	3	2	2	2	3	6	3	
1x	3	3	3	3	3	2	2	2	3	5	3	
2a	3	3	1	6	2	4	3	2	2	2	2	
2i	3	3	1	4	2	4	2	2	3	2	2	
2qu	6	3	2	3	3	3	3	2	2	2	2	
2e	4	3	2	3	3	3	2	2	2	4	3	
2v	6	4	3	3	3	3	2	2	2	4	3	
2x	4	2	3	3	3	3	2	2	3	6	3	
2y	9	1	4	3	3	3	2	2	2	1	4	
2z	2	1	3	2	1	2	2	2	3	4	3	
2ad	3	3	3	3	4	3	2	2	3	3	3	
1d	4	3	7	4	2	3	3	2	3	6	3	b0
1f	3	1	7	4	2	3	3	2	3	4	3	
1g	4	7	7	4	2	3	3	2	3	6	3	
1h	4	7	7	4	2	3	3	2	3	6	3	
1i	5	6	1	5	3	3	2	2	3	4	3	
1l	3	3	3	3	3	3	2	2	3	3	3	
1qu	4	7	3	3	3	3	2	2	3	5	3	
1t	4	4	7	3	3	3	2	2	3	5	3	
1s	3	7	3	3	3	3	2	2	3	5	3	
1ab	6	4	7	4	3	3	2	2	3	6	3	
1ae	3	7	3	3	3	3	2	2	3	5	3	
2d	3	1	7	4	2	3	3	2	3	6	3	
2e	4	7	7	4	2	3	3	2	3	6	3	
2n	8	2	7	4	2	3	3	2	3	6	3	
2ac	12	3	6	5	4	3	3	2	3	6	3	
2ae	2	1	7	3	2	3	3	2	3	4	3	
1a	3	6	3	1	4	3	3	2	3	4	3	Bg
1b	7	4	4	3	3	3	3	2	3	4	3	
1u	10	2	1	1	2	3	3	2	3	4	3	
1w	13	2	1	2	3	3	3	2	3	5	3	
1y	13	3	3	3	3	3	3	2	3	4	3	
1aa	8	4	2	4	3	4	3	2	3	5	3	
2b	7	3	3	3	3	3	3	2	3	4	3	
2g	9	3	3	3	3	3	3	2	3	5	3	
2h	12	3	4	3	3	3	3	2	3	5	3	
2c	10	4	2	2	2	3	3	2	3	4	3	
2p	10	3	3	3	3	3	3	2	3	4	3	
2r	15	3	3	3	3	3	3	2	3	4	3	
2u	10	3	3	3	3	3	3	2	3	4	3	
2ab	12	2	1	1	3	3	3	2	3	4	3	
1e	7	7	7	3	3	3	3	2	3	4	3	BG
1k	18	3	4	2	1	2	2	2	3	5	3	
1m	2	11	3	3	3	3	3	2	3	5	3	
1n	10	6	3	3	3	3	3	2	3	5	3	
1ao	10	2	4	1	1	2	2	2	3	6	3	
1ad	11	1	3	2	2	2	2	2	3	4	3	
2o	11	3	2	2	2	2	2	2	3	4	3	
2f	10	8	4	2	3	3	3	2	3	4	3	
2k	7	4	9	4	3	3	3	2	3	5	3	
2l	10	7	4	4	3	3	3	2	3	5	3	
2m	11	6	3	4	3	4	3	2	3	5	3	
2t	10	7	3	3	3	3	3	2	3	4	3	
2w	10	7	5	3	3	3	3	2	3	4	3	
2ae	11	10	4	2	3	3	3	2	3	4	3	

The division into four groups tried by Correns cannot be considered to be a sharp one. Three plants, 1c, 1p and 1x, used as pollen parents, are classified into the same class (bg); 1x, however, shows a marked difference from 1c



TABLE 2

CORRENS on *Linaria vulgaris*, 1916

		males													
		B	A	C	D	G	H	I	M	E	F	K	L	N	
	B	-	+	+	+	+	+	+	+	+	+	+	+	+	
	A	+	-	-	-	+	+	+	+	+	+	+	+	+	
	C	+	-	-	-	+	+	+	+	+	+	+	+	+	
f	D	+	-	-	-	+	+	+	+	+	+	+	+	+	
e	G	+	+	+	+	-	-	-	-	+	+	+	+	+	
m	H	+	+	+	+	-	-	-	-	+	+	+	+	+	
a	I	+	+	+	+	-	-	-	-	+	?	+	+	-	
l	M	+	+	+	+	-	-	-	-	+	+	+	+	+	
e	E	+	-	-	-	+	+	+	+	-	-	-	-	-	
s	F	+	-	-	-	+	+	+	+	-	-	-	-	-	
.	K	+	-	-	-	+	+	+	+	-	-	-	-	-	
	L	+	-	-	-	+	+	+	+	-	-	-	-	-	
	N	+	-	-	-	+	+	+	+	-	-	-	-	-	

## Theoretical explanation

Parents  $s_1s_2$  and  $s_3s_4$  $F_1$ -generation  $s_1s_3 + s_1s_4 + s_2s_3 + s_2s_4$  $s_1$ -pollen incompatible in  $s_1$ - and in  $s_4$ -styles

		males				
		$s_1s_3$	$s_1s_4$	$s_2s_3$	$s_2s_4$	
f	$s_1s_3$	-	+	+	+	
e	$s_1s_4$	+	-	+	+	
a	$s_2s_3$	+	+	-	+	
l	$s_2s_4$	+	-	+	-	

and from 1p in its behavior towards the other plants of group *bg*, as also towards those of group *bG*. In the same way plants 2b and 2u, ranged in the same class, are different in their behavior towards group *bG*. It is obvious that we are justified to conclude to a rather important difference between some individuals belonging to the same class, so that the composition of this  $F_1$  generation was more complicated than Correns assumed it to be.

A few years after this publication by Correns on *Cardamine*, a brief communication followed (1916) of some results obtained with *Linaria vulgaris*. These results were somewhat different from those with *Cardamine*; the  $F_1$  generation, posterity of one pair of individuals, seemed to consist of 4 groups, each containing plants only that were cross-incompatible with each other, but a difference was found between two reciprocal crosses: one of the groups as a pollen parent was compatible with another group; as seed parent, however, the same group was incompatible to the pollen of the other. This observation of Correns though of a very great importance for the explanation of cross-incompatibility, has been somewhat neglected by the other authors on the same subject.

An analogous division into groups, containing the whole posterity of one pair of parents, was found in later years by other geneticists also. To East and his collaborators, Park and Anderson, we are indebted for a number of careful experiments with individuals of *Nicotiana alata*, *N. commutata* and *N. Forgetiana* and their hybrids. They concluded that the individuals of the  $F_2$  and following generations, issued from two self-incompatible plants, may be grouped into various classes; the number of these classes in the  $F_2$  generation was estimated to be between 12 and 25, a supposition based upon the total percentage of self- and cross-incompatibility, which was calculated to be 15.8. This total incompatibility in the  $F_3$  generation amounts to 19.1 per cent, in the  $F_4$  generation to 26.2 per cent and still higher in the  $F_5$  generation. This increase of total incompatibility is connected with a decrease of the number of classes into which the involved generation can be divided. This calculation of the percentages of incompatibility by East seems to me, on grounds that cannot be discussed here, somewhat inexact, and besides I should like to remark that East's conclusions are based on an assumed identity of reciprocal crosses, an assumption of rather suspect nature. Nevertheless, I can agree with him that the succeeding generations are showing a certain division into classes and that the number of these groups decreases continually, anyhow becomes more and more clear. The cross of *Nicotiana commutata* with *N. Forgetiana* seems to behave in a more simple way; its  $F_1$  generation divided already from the beginning into five distinguishable groups, one of which was formed by one individual only, that was compatible to all other plants and even self-compatible. Especially convincing is the last publication by Anderson (1923), who took advantage of the end-season self-compatibility (pseudo-fertility) occurring rather frequently among the cultures of *Nicotiana* and who obtained thereby families consisting of individuals which by

crosses between them always produced an offspring consisting of two groups only.

Another striking example of the occurrence of intra-incompatible, inter-compatible groups can be found in the work of Lehmann (1919, 1922), on *Veronica syriaca*. The  $F_1$  generation from the two self-incompatible plants used as parents, consisted of four groups, A, B, C and D; within these groups a distinct cross-incompatibility prevailed, but crosses between individuals, belonging to different classes produced always a successful result. Such an inter-cross between two groups ( $B \times D$  for instance) yielded an  $F_2$  genera-

TABLE 3

LEHMANN on *Veronica syriaca*, 1922.p.165. Table I $F_1$ -generation

+ successful pollinations

- unsuccessful pollinations

		males																							
		1	2	3	5	11	17	23	26	1	8	18	24	1	6	9	10	13	22	25	1	4	14	21	1
females	A	21	-	-	-	-	-	-	-	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
		31	-	-	-	-	-	-	-	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
		51	-	-	-	-	-	-	-	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
		111	-	-	-	-	-	-	-	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
		171	-	-	-	-	-	-	-	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
		231	-	-	-	-	-	-	-	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
	261	-	-	-	-	-	-	-	-	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	
B		11	+	+	+	+	+	+	+	-	-	-	-	+	+	+	+	+	+	+	+	+	+	+	+
		81	+	+	+	+	+	+	+	-	-	-	-	+	+	+	+	+	+	+	+	+	+	+	+
		181	+	+	+	+	+	+	+	-	-	-	-	+	+	+	+	+	+	+	+	+	+	+	+
		241	+	+	+	+	+	+	+	-	-	-	-	+	+	+	+	+	+	+	+	+	+	+	+
C		61	+	+	+	+	+	+	+	+	+	+	+	-	-	-	-	-	-	-	+	+	+	+	+
		91	+	+	+	+	+	+	+	+	+	+	+	-	-	-	-	-	-	-	+	+	+	+	+
		101	+	+	+	+	+	+	+	+	+	+	+	-	-	-	-	-	-	-	+	+	+	+	+
		131	+	+	+	+	+	+	+	+	+	+	+	+	-	-	-	-	-	-	+	+	+	+	+
		221	+	+	+	+	+	+	+	+	+	+	+	+	-	-	-	-	-	-	+	+	+	+	+
D		251	+	+	+	+	+	+	+	+	+	+	+	-	-	-	-	-	-	-	+	+	+	+	+
		41	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	-	-	-	-	-
		141	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	-	-	-	-	-
	211	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	-	-	-	-	

Parents  $s_1s_2$  and  $s_3s_4$  $F_1$ -generation  $s_1s_3; s_1s_4; s_2s_3$  and  $s_2s_4$ 

		$s_1s_3$	$s_1s_4$	$s_2s_3$	$s_2s_4$
A	$s_1s_3$	-	+	+	+
D	$s_1s_4$	+	-	+	+
C	$s_2s_3$	+	+	-	+
B	$s_2s_4$	+	+	+	-

tion, consisting of two groups only, which were intra-incompatible, but inter-compatible. This was not only the case in the cross  $B \times D$  but also in the crosses  $A \times C$ , in  $A \times D$  and in  $B \times C$ . So the total  $F_2$  generation grown seemed to consist of eight groups, but not all these groups showed to be different from each other. Cross ( $A \times C$ ), group I, was incompatible when crossed with ( $B \times D$ ), group I; ( $A \times D$ ), group II, incompatible with ( $B \times D$ ), group II, and ( $B \times C$ ), group II, incompatible with ( $A \times D$ ), group I. A number of identical groups seems thus to have been present among these eight classes, found in the various  $F_2$  generations.

As last examples of a typical division into groups, observed by other workers, the results of Baur (1919), Wada (1923) and Terao (1923) may be mentioned. Baur's cross between two self-incompatible individuals of *Antirrhinum hispanicum* resulted into an  $F_1$  generation, consisted of two groups: the individuals belonging to one of these groups were compatible with one of the parents only, those of the other class with both parents. Besides all the individuals were pollenized with pollen of four of their sibs; those of the first group were compatible with three of them and incompatible

TABLE 4

BAUR on *Antirrhinum hispanicum*, 1922  
Tabelle I.p.50

		males					
		EII	EV	101	106	108	110
G	61	+	-	-	-	-	+
r	63	+	-	-	-	-	+
o	65	+	-	-	-	-	+
u	68	+	-	-	-	-	+
p	72	+	-	-	-	-	+
	81	+	-	-	-	-	+
A	82	+	-	-	-	-	+
	84	+	-	-	-	-	+
	88	+	-	-	-	-	+
	94	+	-	-	-	-	+
f	102	+	-	-	-	-	+
e	106	+	-	-	-	-	+
m	108	+	-	-	-	-	+
a	104	+	+	+	+	+	+
l	104	+	-	-	-	-	+
e	67	+	+	+	+	+	-
s	70	+	+	+	+	+	+
	71	+	+	+	+	+	-
	73	+	+	+	+	+	-
	74	+	+	+	+	+	-
G	75	+	+	+	+	+	-
r	76	+	+	+	+	+	-
o	89	+	+	+	+	+	-
u	98	+	+	+	+	+	-
p	105	+	+	+	+	+	-
	107	+	+	+	+	+	-
B	109	+	+	+	+	+	-
	90	+	+	+	+	-	-
	93	+	-	-	+	+	-
	100	+	+	+	-	-	+
	101	+	-	-	-	-	+
	110	+	+	-	+	+	-

Theoretical explanation

Female parent EII  $s_1 s_2$

Male parent EV  $s_2 s_3$

$F_1$ -generation  $s_1 s_3 + s_2 s_3$

		males			
		EII	EV	B	A
		$s_1 s_2$	$s_2 s_3$	$s_1 s_3$	$s_2 s_3$
f	B	+	+	-	+
e	$s_1 s_3$	+	+	-	+
m	A	+	-	+	-
a	$s_2 s_3$	+	-	+	-

" ) Probable printing error in Baur's paper.



to the fourth; the plants of the second group showed an inverse behavior. There were some obvious exceptions, producing some irregular results, but the great majority of the  $F_1$  individuals gave no difficulty in their classification.

A marked contrast to these clear results obtained by various workers is found in the data resulted from the researches of Heribert Nilsson (1916) with rye and Stout (1916 and later) with *Cichorium Intybus* and some other species. Nilsson worked on another line; he did not cross his plants in a systematical way, but he confined his work to an inbreeding of more or less self-compatible individuals and to the statement that the percentages of compatibility in some succeeding generations, produced by using the most favorable methods of isolation, were rather variable. Plants with a 10 to 20 per cent of successful self-pollinations yielded in some cases an offspring of wholly self-incompatible plants and inversely from plants of a very low percentage an offspring was grown with a marked compatibility, giving 20 per cent of grains.

In the very extensive experiments of Stout who worked with systematical crosses, a regularity as found by the other workers was also absent. Stout stated, therefore: "The indisputable evidence, that compatibility and incompatibility in many species are highly variable both in expression and in heredity." And we are obliged to admit that the very rich documentation in Stout's paper does not show any line of regularity, any division in groups. Before all others, however, Stout's observation is of importance, that in many cases reciprocal crosses do not behave in the same way.

TABLE 5

SIRKS on *Verbascum phoeniceum*, 1917 and 1926

F<sub>4</sub>-generation, Family 1918.321

- + successful pollinations
- unsuccessful pollinations
- s slight success

[illegible]

Such an absence of regularity in the behavior of the offspring from one pair of parents was originally stated by my own researches with *Verbascum phoeniceum* (1917); the  $F_1$  generation showed a very complicated behavior; it allowed some preliminary conclusions only: that reciprocal crosses may produce very important differences, that a considerable percentage of cross-incompatibility occurred and that some plants in their conduct towards the other individuals of this  $F_1$  generation seemed to possess a marked female,

TABLE 6

SIRKS on *Verbascum phoeniceum*, 1917 and 1926 $F_5$ -generation, Family 1920.547

+ successful pollinations  
- unsuccessful pollinations  
s slight success

		males																		
		A	B	5	7	10	12	14	6	11	18	2	8	9	13	15	17	20	1	3
female	5	+	+	-	-	-	-	-	+	+	+	+	+	+	+	+	+	+	+	+
	7	+	+	-	-	-	-	-	+	+	+	+	+	+	+	+	+	+	+	+
	10	+	+	-	-	-	-	-	+	+	+	+	+	+	+	+	+	+	+	+
	12	+	+	-	-	-	-	-	+	+	+	+	+	+	+	+	+	+	+	+
	14	+	+	-	-	-	-	-	+	+	+	+	+	+	+	+	+	+	+	+
	6	+	+	+	+	+	+	+	-	-	-	+	+	+	+	+	+	+	+	+
	11	+	+	+	+	+	+	+	-	-	-	+	+	+	+	+	+	+	+	+
	18	+	+	+	+	+	+	+	-	-	-	+	+	+	+	+	+	+	+	+
	2	-	+	-	-	-	-	-	+	+	+	-	-	-	-	-	-	-	+	+
	8	-	+	-	-	-	-	-	+	+	+	-	-	-	-	-	-	-	+	+
male	9	-	+	-	-	-	-	-	+	+	+	-	-	-	-	-	-	-	+	+
	13	-	+	-	-	-	-	-	+	+	+	-	-	-	-	-	-	-	+	+
	16	-	+	-	-	-	-	-	+	+	+	-	-	-	-	-	-	-	+	+
	17	-	+	-	-	-	-	-	+	+	+	-	-	-	-	-	-	-	+	+
	20	-	+	-	-	-	-	-	+	+	+	-	-	-	-	-	-	-	+	+
	1	-	+	+	+	+	+	+	-	-	+	+	+	+	+	+	+	+	-	-
	3	-	+	+	+	+	+	+	-	-	+	+	+	+	+	+	+	+	-	-
	4	-	+	+	+	+	+	+	-	-	+	+	+	+	+	+	+	+	-	-
	15	-	+	+	+	+	+	+	-	-	+	+	+	+	+	+	+	+	-	-
	19	-	+	+	+	+	+	+	-	-	+	+	+	+	+	+	+	+	-	-

Theoretical explanation

 $F_5$ -Parents  $s_1s_2$  and  $as_3s_4$  $F_6$ -generation  $s_1s_3 + s_1s_4 + s_2s_3 + s_2s_4$  $s_1$ -pollen incompatible in  $s_1$ - and in  $s_2$ -styles

		males							
		A	B	5	6	2	1		
		$s_1s_2$	$s_3s_4$	$s_1s_3$	$s_1s_4$	$s_2s_3$	$s_2s_4$		
female	$s_1s_3$	+	+	-	+	+	+	5 etc.	
	$s_1s_4$	+	+	+	-	+	+	6 etc.	
	$s_2s_3$	-	+	-	+	-	+	2 etc.	
	$s_2s_4$	-	+	+	-	+	-	1 etc.	

others a distinct male character, while the remainder formed a series of gradations between these extremes. In the course of seven years since this publication, which was published in 1917, the work was continued on a more or less extensive scale and the results were in the three succeeding generations the same, but from the  $F_4$  generation on a somewhat more distinct regularity could be observed. Among the individuals of this  $F_4$  generation

TABLE 7

SIRKS on *Verbascum phoeniceum*, 1917 and 1926

$F_6$ -generation. Family 1920.549

+ successful pollinations  
- unsuccessful pollinations  
s slight success

		males															
		A	B	8	13	14	15	20	2	9	10	18	19	1	6	12	17
females	8	+	+	-	-	-	-	-	+	+	+	+	+	+	+	+	+
	13	+	+	+	-	-	-	-	+	+	+	+	+	+	+	+	+
	14	+	+	+	-	-	-	-	+	+	+	+	+	+	+	+	+
	15	+	+	+	-	-	-	-	+	+	+	+	+	+	+	+	+
	20	+	+	+	-	-	-	-	+	+	+	+	+	+	+	+	+
	2	+	-	-	-	-	-	-	-	-	-	-	-	+	+	+	+
	9	+	-	-	-	-	-	-	-	-	-	-	-	+	+	+	+
	10	+	-	-	-	-	-	-	-	-	-	-	-	+	+	+	+
	18	+	-	-	-	-	-	-	-	-	-	-	-	+	+	+	+
	19	+	-	-	s	-	-	-	-	-	-	-	-	+	s	+	+
	1	-	+	-	-	-	-	-	+	+	+	+	+	-	-	-	+
	6	-	+	-	-	-	-	-	+	+	+	+	+	-	-	-	+
	12	-	+	-	-	-	-	-	+	+	+	+	+	-	s	-	+
	17	-	+	-	-	-	-	-	+	+	+	+	+	-	-	-	+
	3	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
	4	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
	5	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
	7	-	-	-	-	-	-	-	-	-	-	-	-	-	s	-	-
	11	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
	16	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-

Theoretical explanation

$F_5$ -Parents  $s_1s_2$  and  $s_3s_4$

$F_6$ -generation  $s_1s_3 + s_1s_4 + s_2s_3 + s_2s_4$

$s_1$ -pollen incompatible in  $s_1$ - and in  $s_2$ -styles

$s_3$ -pollen incompatible in  $s_3$ - and in  $s_4$ -styles

		males							
		A	B	8	2	1	3		
		$s_1s_2$	$s_3s_4$	$s_1s_3$	$s_1s_4$	$s_2s_3$	$s_2s_4$		
females	$s_1s_3$	+	+	-	+	+	+	8 etc.	
	$s_1s_4$	+	-	-	-	+	+	2 etc.	
	$s_2s_3$	-	+	-	+	-	+	1 etc.	
	$s_2s_4$	-	-	-	-	-	-	3 etc.	

some plants were found, which behaved themselves towards their sibs wholly identical. In the  $F_4$  family 1918, 321 for instance, the 20 individuals grown, contained one small group of 3 individuals, that showed the same way of reaction versus the other 17 plants, and another group of 2 individuals. The remaining 15 plants could be divided into two classes according to their behavior towards these two groups, but within these two classes no regularity seemed to be present.

It was therefore a rather unexpected result, when I obtained in the sixth generation two families, each consisting of 20 plants, which showed a distinct classification into 4 groups. One of these families, 1920, 547, was classified into four classes, wholly intra-incompatible and among themselves compatible; two combinations, however, excepted. Group III female crossed with group I male gave no results, while the reciprocal crosses were all compatible. The same was found in the cross group IV female with group II male; in this direction the crosses were incompatible. The previously stated difference in reciprocal behavior between certain individuals was now observed to be present between whole groups, and the phenomenon must therefore be considered of a fundamental importance.

This same phenomenon asserted itself stronger still in another family of the  $F_6$  generation, 1920, 549. Here the four distinguishable groups were intra-incompatible too, but in reciprocal crossing only one of the six combinations proved to be successful in both directions, so that the difference between the reciprocal crossings in this family was indeed very striking. Group I was as female parents compatible with all three others, as male parent, however, incompatible; group II as male compatible with two of them, as female also; group III showed the same behavior: compatible with two, incompatible with two; while group IV as female was incompatible with the other three, as male compatible to them. So it is very probable, that in consequence of the continuous sister-brother-mating from the confusing mass of types, which there appeared to exist in the  $F_1$  generation, only a few of them have been kept, which are of a much simpler construction and which consequently show a much greater regularity with regard to their behavior in reciprocal crossings.

A peculiar analogy to these phenomena of self- and cross-incompatibility among higher plants was found by Kniep with Basidiomycetes, for instance with *Schizophyllum commune*. In a series of very beautiful researches with fungi with regard to what he considers as phenomena of sexuality, he comes first of all to an affirmation of Blakeslee's earlier work on plus—and minus—races, which are said to represent the two sexes in the haploid stadia of various fungi. But the experiments with *Schizophyllum* led to more complicated results. When the mycelia proceeding from the spores of one single fruit-body are systematically mated, the mycelia prove to belong not only to two groups, male and female, but they prove to form four groups, which are all intra-incompatible, which means that they do not copulate when combined within the group and are only compatible with the three other groups. In some families there happen to be, apart from the four groups, some single



individuals still, which can copulate with two of the formed groups, and which consequently are more or less exceptions.

The analogy with the numerous formations of groups among the offspring of a pair of self-incompatible plants is evident, but should on the other hand, not be valued too highly; with fungi we had to do with haploid mycelia, with higher plants with a process between the style, an organ of diploid character and the pollen tube, that may be considered probably as haploid.

Summarizing what is said before, we get a strong indication that the formation of distinct groups in the progeny of a pair of self-incompatible individuals is regular, and that consequently this formation of groups must be founded on certain existing genotypical factors.

The formation of groups did not appear in all researches, it is true, so as in Stout's experiments on *Cichorium*, but that they present themselves in numerous other experiments of various workers, and lastly also in my *Verbascum* cultures, may prove sufficiently that they are of importance and that consequently the foundation of self-incompatibility is a genotypical one. It is another question, however, to what extent real Mendelian factors are at play here. Correns, East, Lehmann have tried many times to epitomize their results in Mendelian formulas, but their efforts had either no complete success, or had only significance for their own data with neglect of those of the other workers. After that, in 1921, Prell suggested a solution founded on the data of literature, which is rather well in accord with all the facts, and which, with some alteration and extension, is sure to gain a further stretching meaning too. Prell's hypothesis has recently (1925) been adopted by East and Mangelsdorf, and has the following leading thought: In every diploid organism, which is self-incompatible, there are two oppositional factors, let us say  $s_1$  and  $s_2$ , which in the style inhibit the growth of pollen tubes possessing one of these factors. Consequently, flowers from a plant with the formula  $s_1s_2$  can only be fertilized by pollen grains which either do not possess any  $s$ -factors at all, or if they do so, others than  $s_1$  or  $s_2$ , so for instance  $s_3$  or  $s_4$ . Consequently self-fertilization is quite out of question and cross-fertilization only possible with organisms that are not also  $s_1s_2$ . First condition is, that selection between the various pollen grains is possible in the fertilization, an  $s_1s_2$  plant can for instance be crossed with an  $s_2s_3$  individual, but only by the  $s_3$  pollen grains, while the  $s_2$  pollen is inhibited in its growth through the style and remains unsuccessful. Selective fertilization, or as we in Europe call it on instigation of Heribert-Nilsson, certation between pollen grains is taking place and playing an important part in the process of self- and cross-incompatibility. Most results of the discussed researches with only one exception may be formulated in the way of this hypothesis of oppositional factors; those of Correns are rather difficult because of the want of complete combination of all the  $F_1$  individuals among each other and because of the contradictory facts in his results, but as the formulas, added to each of the schemes show, in all cases a solution is possible on the lines of the hypothesis of Prell, East and Mangelsdorf.

A slight extension, however, of this hypothesis is necessary; the hypothesis is not sufficient for an explanation of the difference between the reciprocal crosses, and these differences may now be considered to exist really. East has denied them and has founded his previous explanation on the identity of reciprocal crosses. In his more recent publication (1925), however, he accepts them. Lehmann did not find them. Correns showed them to be present in *Linaria*, Stout in *Cichorium* and my own researches in *Verbascum*. In order to explain these phenomena, East thinks that he may suppose that for instance in an  $s_1s_2$  plant there can by way of exception also grow  $s_1$  pollen, from which homozygous  $s_1s_1$  plants are produced. Such  $s_1s_1$  plants crossed with  $s_1s_2$  individuals show a difference in reciprocity;  $s_1s_1$  as male with  $s_1s_2$  as female is incompatible, inversely the crossing is compatible.

In my opinion there is, however, another solution which is possible also; namely the supposition that for instance  $s_1$  pollen is incompatible not only in an  $s_1$  style, but also in an  $s_2$  style, while inversely  $s_2$  pollen can function in an  $s_1$  style.

It may therefore be considered a fact, that in self-incompatible plants there are genotypical factors, causing the incompatibility of pollen grains containing one of these factors.

But a second problem is, if these factors are fixed and unalterable. Stout's

TABLE 8  
PRELL, 1921—EAST AND MANGELSDORF, 1925  
HYPOTHESIS OF OPPOSITIONAL FACTORS

	MALES					
	$s_1s_1$	$s_2s_2$	$s_3s_3$	$s_1s_2$	$s_1s_3$	$s_2s_3$
$s_1s_1$	-	+	+	+	+	+
$s_2s_2$	+	-	+	+	+	+
$s_3s_3$	+	+	-	+	+	+
$s_1s_2$	-	-	+	-	+	+
$s_1s_3$	-	+	-	+	-	+
$s_2s_3$	+	-	-	+	+	-

results, those of Heribert-Nilsson, East's original data with *Nicotiana* and my own in *Verbascum* show that the regularity discussed here is not always present, anyway not from the very beginning. Close inbreeding, however, seems to be the way of bringing it about. This absence of any regularity, too, must be accounted for. Up to now the data on that subject are still too scarce, but one is inclined to assume that the solution will have to be found on the line of Goldschmidt's view of quantitative inheritance. The most probable supposition is that the self-incompatible plants, considered as species, contain a number of oppositional factors as multiple allelomorphs, partly different in quantity, partly probably in quality.

Kniep also has taken this view with regard to his experiments on Basidiomycetes, and with the help of supposed mutations in these factors, he has also accounted for the exceptions he had found. On this line our knowledge is still too incomplete, than that a well-founded decision should be possible.

The interpretation of the greater complexity in the first generations may be thought in two directions: a series of multiple allelomorphs or a number of multiple factors, the inheritance of which is independent. In the last assumption we are obliged to accept the occurrence of frequent mutations by loss, so that in the later generations one pair of oppositional factors only is left; assuming a series of multiple allelomorphs we must suppose the possibility that one pair of such factors can produce a number of different allelomorphs. Personally I prefer this last supposition, but I will not discuss the matter here as I hope to do so in a paper before the section of genetics at the International Congress on Plant Sciences at Ithaca.

Quantitative differences of these factors are not at all improbable if we look for their physiological nature on the line of differences in concentration, as was done by me in 1917, and later also by Tischler (1918), von Ubisch (1923) and Ernst (1925) for the explanation of the phenomena of legitimate and illegitimate pollination in heterostyle plants, whether or not coinciding with more or less self-incompatibility.

However, as matters are now, such a physiological interpretation of facts, which unquestionably goes further into the matter than a purely genotypical one, is not yet possible. In the researches into that question, special attention will have to be paid to the intensity of self-incompatibility as it shows itself in the works of Heribert-Nilsson and Stout; very probably the plants, that are best fitted for that purpose will be those with only a few seeds in each fruit, so as for instance the races of *Brassica*.

A question of secondary importance is, in how far the phenomena of self-incompatibility should be considered phenomena of sexuality; Correns (1926) considered Kniep's results with *Schizophyllum* as phenomena of incompatibility; Kniep as pluripolar sexuality.

I do not think it desirable to discuss this subject here; for the present that sort of contemplations are too subjective and degenerate only too often in an altercation of words, which I would rather avoid.

Lastly there still remains the problem of the relation between self-compatible and self-incompatible plants. I cannot discuss any researches of my own on this subject; in my experiments with *Verbascum* I never obtained self-compatible individuals. But in the work of others there are to be found

TABLE 9  
POSSIBLE EXPLANATION OF CROSS  
SELF-COMPATIBLE x SELF-INCOMPATIBLE

COMPTON

Incompatible parent  $s_1s_1$

Compatible parent  $s_0s_0$

$F_1$ -generation  $s_1s_0$

$s_1$ -pollen compatible in  $s_1s_0$ , but not in  $s_1s_1$

		pollen	
		$s_1$	$s_0$
e g s	$s_1$ 1)	$s_1s_1$	3) $s_1s_0$
	$s_0$ 2)	$s_0s_1$	4) $s_0s_0$
$F_2$		{ 3 compatible ( 2, 3, 4 ) 1 incompatible ( 1 ).	

EAST

Incompatible parent  $s_1s_1 \cdot s_2s_2$

Compatible parent  $s_0s_0 \cdot s_0s_0$

$F_1$ -generation  $s_1s_0 \cdot s_2s_0$

$s_1s_2$ -pollen incompatible;

$s_1s_0$  and  $s_0s_2$  compatible, but with diminished rapidity of growth

		pollen			
		$s_1s_2$	$s_1s_0$	$s_0s_2$	$s_0s_0$
rapidity of growth		0	1	1	3
e g s	$s_1s_2$ 1)	-----	5) $s_1s_2 \cdot s_1s_0$	9) $s_1s_2 \cdot s_0s_2$	13) $s_1s_2 \cdot s_0s_0$
	$s_1s_0$ 2)	-----	6) $s_1s_0 \cdot s_1s_0$	10) $s_1s_0 \cdot s_0s_2$	14) $s_1s_0 \cdot s_0s_0$
	$s_0s_2$ 3)	-----	7) $s_0s_2 \cdot s_1s_0$	11) $s_0s_2 \cdot s_0s_2$	15) $s_0s_2 \cdot s_0s_0$
	$s_0s_0$ 4)	-----	8) $s_0s_0 \cdot s_1s_0$	12) $s_0s_0 \cdot s_0s_2$	16) $s_0s_0 \cdot s_0s_0$

$F_2$	16 compatible:	7	8	10	12	13	14	15	16	Total
	4 incompatible:	5	6	9	11	Total				
		1	1	1	1	4				

The numbers 6 and 11 (incompatible) will show probably a greater "pseudofertility" than 5 and 9, owing to the fewer number of  $s_1$ - and  $s_2$ -factors.



some data on this problem. Firstly the results of Compton (1912, 1913) with *Reseda*, East (1919) with *Nicotiana* and Baur (1919) with *Antirrhinum*, who crossed self-compatible individuals with self-incompatible ones. All these three workers agree in the conclusion that self-compatibility must be considered a dominant character opposed to self-incompatibility as a recessive one. The  $F_1$  of their crossings was self-compatible indeed, while the  $F_2$  generation segregated into self-compatible and self-incompatible individuals. Compton did not publish any numbers, but was speaking only of a ratio of 3:1; East found 144:37 or 3.89:1, while Baur obtained 190 self-compatible versus 22 self-incompatible individuals, or 8.64:1. On the other hand we have the fact that in various cultures self-compatible plants were found as descendants from really self-incompatible ones. East ascribes this appearance to an error of experiment, it is true, but Correns found it sporadically in *Cardamine*, Stout rather frequently in *Cichorium*, *Eschscholtzia*, *Verbascum* and others, Heribert-Nilsson in rye.

Yet, in my opinion, these two results need not to be contradictory to each other if we may accept what has been demonstrated in what precedes, that incompatibility is the consequence of oppositional factors and that in self-compatible plants these factors are absent, then it is self-evident that there can be no question of a real dominance of compatibility, while yet the  $F_1$  generation is compatible.

Indeed we can represent all this in formulas in this way: Parents incompatible  $s_1s_2 \times$  compatible  $s_0s_0$ .  $F_1$  generation,  $s_1s_0$  and  $s_2s_0$ .

So these  $F_1$  plants are self-compatible in consequence of the presence of  $s_0$  pollen, while we may suppose besides that  $s_1$  pollen can grow in the heterozygous style as well, be it then more slowly. The ratios found in the  $F_2$  generation are then the result of the proportions in the rapidity of growth between  $s_0$  and  $s_1$  pollen in the quantity of pollen, which has been used for the pollination.

On the other hand it is then very well possible that from a population of self-incompatible plants, there are individuals isolated, which do not possess any oppositional factors, and which are consequently self-compatible.

As the schemes here will show, the data of Compton and East are in very good accord with this view; Baur's results only are more complicated and not sufficient to warrant any conclusions.

Firstly, in the cross of *Antirrhinum hispanicum* with *A. majus peloricum*, which he has studied, there enters an important percentage of true sterility of the gametes, and, secondly, the ratios in the four  $F_1$  families grown diverge rather strongly. That, however, for the segregation zygomorph versus peloric there has been found a ratio differing so largely from what we had expected 3:1, may very probably be ascribed to a linkage between the factor for incompatibility and the factor for the zygomorphic form of the flowers.

Important in this regard are still the greatly different grades of self-incompatibility which came to light in the  $F_2$  generations of Baur and East,

and the strong end-season self-compatibility appearing in East's hybrids. This surely indicates a great number of multiple allelomorphs playing a part here.

Summarizing we are, through the researches from American and European sides with regard to the genotypical problems of self- and cross-incompatibility much better informed than we were ten years ago; but I hope that I have demonstrated that still many unsolved problems remain hidden under the complex of horticultural, genotypical and physiological problems, so utterly important and attractive both for science and practice.

Since the above paper was written, a study by Filzer (1926) from Lehmann's laboratory was published in which the writer is founding an interpretation of his experiments on the same assumptions as those of Prell, East and Mangelsdorf.

### LITERATURE CITED

- Anderson, E., 1924. Studies on self-sterility. VI. The genetic basis of cross-sterility in *Nicotiana*. *Genetics* 9: 13-40.
- Baur, E., 1919. Ueber Selbststerilität und über Kreuzungsversuche einer selbstfertilen und einer selbststerilen Art in der Gattung *Antirrhinum*. *Zeits. Indukt. Abstamm.* 21: 48-52.
- Compton, R. H., 1912. Preliminary note on the inheritance of self-fertility in *Reseda odorata*. *Proc. Cambr. Phil. Soc.* 22: 7.
- 1913. Phenomena and problems of self-sterility. *New Phytologist* 22: 197-206.
- Correns, C., 1913. Selbststerilität und Individualstoffe. *Biolog. Centralblatt* 23: 389-423.
- 1916. Individuen und Individualstoffe. *Die Naturwissensch.* 4: 183-187, 193-198, 210-213.
- 1926. Ueber Fragen der Geschlechtsbestimmung bei höheren Pflanzen. Bericht fünfte Jahresversammlung Deutsche Gesselsch. f. Vererbungswissenschaft, pp. 5-40; also *Zeits. Indukt. Abstamm.* 41: 5-41.
- East, E. M., 1915. An interpretation of self-sterility. *Proc. Nat. Acad. Sci.* 1: 95-100.
- 1915. The phenomenon of self-sterility. *Amer. Nat.* 49: 77-88.
- 1917. The behavior of self-sterile plants. *Science, N. S.* 46: 221-222.
- 1917. The explanation of self-sterility. *Jour. of Heredity* 8: 382-383.
- 1918. Intercrosses between self-sterile plants. *Brooklyn Botanic Garden Memoirs* 1: 141-153.
- 1919. Studies on self-sterility. III. The relation between self-fertile and self-sterile plants. *Genetics* 4: 341-345.
- 1919. Studies on self-sterility. IV. Selective fertilization. *Genetics* 4: 346-355.
- 1919. Studies on self-sterility. V. A family of self-sterile plants, wholly cross-sterile inter se. *Genetics* 4: 356-363.
- 1923. Genetical aspects of self- and cross-sterility. *Am. Jour.* 10: 468-473.
- East, E. M., and Mangelsdorf, A. J., 1925. A new interpretation of the hereditary behavior of self-sterile plants. *Proc. Nat. Acad. Sci.* 9: 166-171.
- East, E. M., and Park, J. B., 1917. Studies on self-sterility. I. The behavior of self-sterile plants. *Genetics* 2: 505-609.
- 1918. Studies on self-sterility. II. Pollen-tube growth. *Genetics* 3: 353-366.
- Ernst, A., 1925. Genetische Studien über Heterostylie bei *Primula*. *Archiv Julius Klaus-Stiftung.* 1: 13-62.
- Filzer, P., 1926. Die Selbststerilität von *Veronica syriaca*. *Zeits. Indukt. Abstamm.* 41: 137-197.
- Heribert-Nilsson, N., 1916. Populationsanalysen und Erblchkeitsversuche über die Selbststerilität, Selbstfertilität und Sterilität bei dem Roggen. *Zschr. f. Pflanzenzüchtung* 4: 1-44.
- Kniep, H., 1919. Ueber die morphologische und physiologische Geschlechts-differenzierung. (Untersuchungen an Basidiomyceten.) *Verhandl. Phys. med. Gesellsch. Würzburg* 46: 1-18.
- 1922. Ueber Geschlechtsbestimmung und Reduktionsteilung. (Untersuchungen an Basidiomyceten.) *Verhandl. Phys. med. Gesellsch. Würzburg* 47: 1-29.
- 1923. Ueber erbliche Aenderungen von Geschlechtsfaktoren bei Pilzen. *Zeits. Indukt. Abstamm.* 31: 170-183.
- Lehmann, E., 1919. Ueber die Selbststerilität von *Veronica syriaca*. *Zeits. Indukt. Abstamm.* 21: 13-40.

- 1922. Ueber die Selbststerilität von *Veronica syriaca*. II. Zeits. Indukt. Abstamm. 27: 161-177.
- Munerati, O., 1923. Beitrag zum Studium der Selbstunempfänglichkeit des Roggens. Zschr. Pflanzenzüchtung 9: 176-178.
- Prell, H., 1921. Das Problem der Unbefruchtbarkeit. Naturwiss. Wochenschr. N. F. 20: 440-446.
- 1921. Anisogametrie, Heterogametrie und Aëthogametrie als biologische Wege zur Förderung der Amphimixis. Archiv f. Entwicklungsmechanik 49: 463-490.
- Sirks, M. J., 1917. Stérilité, auto-inconceptibilité et différenciation sexuelle physiologique. Archives Neerl. Sci. Exactes et Natur. Série IIIB, 3: 205-234.
- 1926. Further data on the self- and cross-incompatibility of *Verbascum phoeniceum*. Genetica 8: 344-367.
- Stout, A. B., 1916. Self- and cross-pollinations in *Cichorium Intybus* with reference to sterility. Mem. New York Bot. Gard. 6: 333-454.
- 1917. Fertility in *Cichorium Intybus*: The sporadic occurrence of self-fertile plants among the progeny of self-sterile plants. Amer. Jour. of Bot. 4: 375-395.
- 1918. Experimental studies of self-incompatibilities in fertilization. Proc. Soc. Exp. Biol. Med. 15: 51-54.
- 1918. Fertility in *Cichorium Intybus*: Self-compatibility and self-incompatibility among the offspring of self-fertile lines of descent. Jour. Genetics 8: 71-103.
- 1920. Further experimental studies on self-incompatibility in hermaphrodite plants. Jour. Genetics 9: 85-129.
- 1922. Cyclic manifestation of sterility in *Brassica pekinensis* and *B. chinensis*. Bot. Gaz. 73: 110-132.
- 1923. The physiology of incompatibilities. Am. Jour. Bot. 10: 459-461.
- Terao, H., 1923. On the inheritance of self-sterility (Jap.). Idengaku Zassi. Jap. Jour. Genetics 2: 144-155.
- Tischler, G., 1918. Analytische und experimentelle Studien zum Heterostylie-Problem bei *Primula*. Festschr. Landw. Hochschule Hohenheim, pp. 254-273.
- 1918. Untersuchungen über den anatomischen Bau der Staubund Fruchtblätter bei *Lythrum Salicaria* mit Beziehung auf das Illegitimitätsproblem. Flora 111: 162-193.
- Ubisch, G. V., 1923. Versuche über Vererbung und Fertilität bei Heterostylie und Blütenfüllung. Zeits. Botanik 15: 193-232.
- Wada, U., 1923. Experiments on the breeding and heredity of sweet potato (Jap.). Idengaku Zassi. Jap. Jour. Genetics 2: 137-144.





# STUDIES OF THE INHERITANCE OF SELF AND CROSS-INCOMPATIBILITY

A. B. STOUT  
New York Botanical Garden  
(WITH PLATES 28-29)

Incompatibilities *within* a species, a variety, or a culture of plants involve a very special kind of physiological condition which discriminates between fertilizations so that certain pollinations result in the production of fruit and seeds while others do not. They are most distinctively exemplified in the self- and cross-incompatibilities of hermaphrodite plants whose bisexual flowers are homomorphic and self-pollinating.

Incompatibilities both self and cross exist in certain species of many families of flowering plants from the grasses to the composites. This, of itself, indicates that there is some very fundamental biological significance in their development and operation. In some genera, as in *Lilium*, self-incompatibilities appear to be present in many, if not in every, species. In other genera, of which *Antirrhinum* and *Nicotiana* may be mentioned, certain species may evidently be free of incompatibilities while in another species they are strongly developed. Such behavior clearly indicates that at least the capacities for manifesting such incompatibilities are in some degree transmissible in heredity. The condition responsible for incompatibilities in fertilization is present rather generally in certain species and is in some way passed on from parents to their progeny. The evidence as to the degree in which such incompatibilities are inherited should be based both on the study of the behavior of individuals and on the study of series of pedigreed cultures.

On the basis of our studies with species of *Lilium*, *Brassica*, *Linaria*, *Cichorium* and *Hemerocallis* it is to be questioned whether, in these species at least, strains of plants which are propagated from seed and which are fully self-incompatible exist in nature or can be isolated by selection. Individual plants which are more or less self-compatible are to be found and such plants continue to appear even among the progeny of parents that are fully self-incompatible.

Of the species *Linaria grandiflora*, all of a series of 16 plants taken at random were found to be fully self-incompatible. From seed of three different crosses among these plants, 204 plants were grown to maturity and tested. Of these, two plants were found to be self-compatible to a rather marked degree.

Four generations of plants originating in a cross between *Linaria dalmatica* and *L. vulgaris* were studied, the parents in each generation being plants fully self-incompatible. The  $F_1$  consisted of three plants which were all fully self-incompatible. In the  $F_2$ , consisting of 173 plants, 19 were self-

compatible in some degree. In the  $F_3$ , consisting of 128 plants, 7 were self-compatible. In the  $F_4$ , consisting of 127 plants, two were self-compatible.

Sixteen plants of *Linaria grandiflora*, comprising a random population, were fully self-incompatible, but of the next generation of 240 plants two were found to be self-compatible.

Results similar to the above have been obtained in several species of *Lilium*, in *Cichorium Intybus* (Stout, 1916 and 1917) and in two species of *Brassica* (Stout, 1922), except that, as a rule, the proportions of plants that were self-compatible were larger. Similar results have been obtained by others who have studied this type of sterility. In these plants the self-compatibility is so frequent and, in some cases at least, so pronounced that they are not to be considered as "pseudo self-fertility."

In the plants I have studied, the highly self-compatible plants set viable seed to self-pollinations made at the proper time for pollination. No premature pollination is necessary. Furthermore no amount of "premature pollination" can induce seed-setting by fully self-incompatible plants. Results typical of these behaviors are shown in A of PLATE 28.

For purposes of broad and rather loose genetical analysis it may be necessary to regard such species as fully self-incompatible. But the fact is that whenever a large number of plants are grown in a pedigreed line some of the plants set fruit with viable seed to self-pollination through quite normal pollinations and true fertilizations. Such plants continue to appear among progenies even after several generations of selection of parents for complete self-incompatibility.

The theory that a pair of hereditary oppositional factors are operating in self- and cross-incompatibility has been reported and demonstrated, especially by Lehmann (1919 and 1922), Filzer (1926), East and Mangelsdorf (1925) and by others. We must assume, on this basis, that in the appearance of self-compatibility there has been a mutation of at least one of these factors such as a change from  $S_1S_2$  to  $S_1S_0$ . Then plants with a genetical complex of  $S_1S_0$  should be self-compatible, at least to pollen carrying  $S_0$ . Evidence as to whether such a mutation has occurred may readily be had from the behavior of the offspring of these self-compatible plants which arise sporadically. The progenies of selfings continued through several generations should be studied. This we have done especially in the species *Cichorium Intybus* and *Brassica pekinensis*, and part of the results have been published (Stout, 1920 and 1922).

Thus far these studies have all yielded the same general result. In the first generation of such plants the number of self-compatible individuals increases decidedly. There is an immediate result which clearly indicates that self-compatibility thus arising is heritable in a certain grade or degree. But continued selection in subsequent generations has not yet given a further appreciable increase in the relative number of self-compatible plants and we have not been able to isolate any strain that continues to be fully self-compatible.

In *Cichorium Intybus* selection for high degrees of self-compatibility was continued in two families through two generations with rather large numbers of plants. In one family the second generation comprised 258 plants of which 100, or two out of five, were self-compatible to some degree. In the other family there were 471 plants in the second generation of which 266, or more than half, were self-compatible to some extent.

In *Brassica pekinensis*, selections for the highest grades of self-compatibility, which in this plant is a mid-period self-compatibility, have now been continued through seven generations during which a grand total of over 5,000 plants were grown and tested. In the very first generation of the main line studied, 44 out of 88 plants were self-compatible to the degree of producing viable seeds and for some plants there was full and complete fertility to selfing for a time during the mid-season of bloom. Thereafter the relative numbers of self-compatible plants varied considerably but in the seventh generation of 556 individuals there were only 385 that were self-compatible to the degree that viable seeds were produced to self-pollination.

TABLE 1

DATA FOR *Brassica pekinensis*: SIX SERIES OF SISTER PLANTS HAVING SEVEN GENERATIONS OF SELFED PARENTS EACH HIGHLY SELF-COMPATIBLE AND ALL DESCENDING FROM ONE SINGLE PARENT

CROP OF 1926 NO. OF SERIES	NUMBER OF PLANTS			
	ENTIRELY SELF- INCOMPATIBLE	SELF-COMPATIBLE		
		FEEBLY	MEDIUM	HIGHLY
141. ....	22	13	12	44
142. ....	18	8	8	26
143. ....	25	5	19	32
144. ....	16	5	21	70
146. ....	9	9	8	74
149. ....	25	16	21	50
	115	56	89	296

In this tabulation, plants that were feebly self-compatible produced small capsules but matured no viable seeds and hence could be classed as self-incompatible as well as feebly self-compatible. Plants that were classed as entirely self-incompatible produced no capsules to self-pollination.

In these studies always the most highly self-compatible plants were selected as parents for the next generation. Sometimes all the plants of a sister series were more or less self-compatible. In several cases this condition continued for two successive generations and it seemed that a line fully self-compatible had been segregated and that the factors for self-incompatibility had been eliminated by the line breeding. If the culture had been discontinued at this stage, such a conclusion would apparently have been justified. But the continued culture of the line by controlled selfings always gave some plants, and often a considerable proportion of them, entirely self-incompatible as is shown in TABLE 1.



I have not as yet found it possible to isolate a strain or pedigreed line consisting of any considerable number of plants which continually breeds true to the character of self-compatibility. If, in the first appearance of self-compatible plants from progeny of self-incompatible parents, there was a stable mutation of one of the oppositional factors as from  $S_1S_2$  to  $S_1S_0$  the first generation from such a plant should give the two classes  $S_1S_0$  and  $S_0S_0$  with all members of each self-compatible at least to 50 per cent of their ability in the mid-period of bloom, and the succeeding generations should have so continued unless further mutation occurred. The fact is that even with repeated selection the condition of  $S_0S_0$  was not obtained in any culture if we are to judge by the breeding tests.

It is clear that, considered as contrasted characters in these species of *Brassica* and *Cichorium*, factors for self-incompatibility have greater value and dominance in expression than have factors for self-compatibility and they show a greater selection value in line breeding. For an assumed factor for self-incompatibility, a value of 1 may thus be closely approximated. As strictly contrasted characters in crosses within such species neither character is actually dominant for the two conditions are reversible to the degree that in line breeding either one may arise or segregate from the other. The value of "S", therefore, continues to shift or "mutate" between 1 and near 0.

It must be recognized also that in these cultures there were many grades or degrees of self-compatibility and self-incompatibility if we are to judge the actual performance of plants by the number of fruits produced, by their size and shape, and by the number of viable seeds they contain. The range from complete self-incompatibility ( $S_1$ ) to the highest grade of self-compatibility usually presents numerous gradations and the greatest degree of the latter is nearly always below and many times far below the possible or potential fertility. This clearly indicates that at no time is the physical basis of self-incompatibility eliminated in the genetical composition of the individual in these species. The condition  $S_0S_0$  is not fully realized although this condition is approximated in many individual plants of the lines selected for self-compatibility. We must, therefore, represent the value of S as fluctuating from a value of 1 toward a value of 0 in various individuals of each generation.

It has been mentioned above that plants which are somewhat self-compatible are usually to be found in so-called self-incompatible species. When such a plant is extensively propagated vegetatively, as in clonal varieties of apples and pears, there is a chance to test the clon under widely different conditions in some of which the precise and more limited conditions for self-fertilization may be met, a result that may never be realized in the experimental testing of an annual plant. There has been much discussion as to whether any clonal apple variety is fully self-incompatible, and certain of the earlier investigators have even expressed the opinion that no plant is absolutely self-incompatible. But among widely cultivated clonal varieties, some, such as the tiger lily (*Lilium tigrinum*) and the orange day lily (called *Hemerocallis fulva*), have never been known to yield a single capsule with



seed to self- and close-pollination. Among annual plants investigated, some species, as *Veronica syriaca*, appear to include no plants in the least degree self-compatible as reported by Lehmann (1919, 1922). Sirks (1917) also finds no self-compatible plants in his cultures of *Verbascum phoenicium*. Such species are evidently the exception. Many so-called self-incompatible species contain some plants more or less self-compatible and fully or continually self-incompatible individuals among perennials appear to be scarce.

In these cases of partial self-incompatibility the influence of any factors or genes for self-incompatibility or self-compatibility may perhaps be at times suspended. Some clew as to how this may occur may be found in the cases in which a shift from self-incompatibility to self-compatibility occurs at a particular period of the flowering cycle. In two species of Chinese cabbages (*Brassica chinensis* and *B. pekinensis*) the plants which are self-compatible are most fully so for those flowers that mature during the mid-period of bloom (Stout, 1922). For such plants a group of the first and of the last flowers are fully self-incompatible and in them the pollen tubes from self-pollination do not penetrate the stigma but merely grow on the surface into a tightly coiled knot. Thus in the first flowers that bloom the  $S_1\bar{S}_2$  reaction is fully expressed. In the flowers that open a few days later on the same inflorescence, self-fertilization may occur and seeds result. Thus in a series of sister flowers on the same plant there is a change from self-incompatibility to full compatibility followed by a return to self-incompatibility. Such a cyclic change clearly demonstrates a biogenetic or developmental control of the assumed factorial organization of the germ plasm. This may be and evidently is a highly specialized condition but it illustrates the possibility of such influence in the differentiation of entire individuals as such. As to the values of such assumed hereditary factors, it is clear that the same germ plasm composition in pistils and in pollen may come to expression at one time in full self-incompatibility and at another time in full self-compatibility. In the behavior of individual flowers of a single inflorescence the condition of  $S_1\bar{S}_2$  is at one time realized while at another time  $S_1S_0$ , or even  $S_0S_0$  is in expression.

In respect to cross-incompatibilities within a series or a culture of plants, the most significant behavior for genetical analysis is that of simple group reactions so graphically presented by various investigators. In the simplest of these cases there are groups of plants that are intra-cross-incompatible but between the various groups there is complete cross-compatibility and in all the reciprocals each pair yields the same result. In these group relations the rule is that when two plants are cross-sterile with a third plant they will be cross-sterile to each other.

From the beginning of my studies of incompatibilities the group reactions have continually failed to conform to this scheme of behavior. In the first place there have been many grades or degrees of compatible and incompatible relations quite as were recognized by Correns (1912, 1913) in one of the earliest studies made of group relations in fertilization. Furthermore my

material has regularly fallen into large rather than small numbers of such groups with each group consisting of a relatively small number of individuals (often only two), and frequently a single plant is included in a number of groups thus breaking the rule of intra-group incompatibility and inter-group compatibility.

TABLE 2

RESULTS OBTAINED WHEN 12 SISTER PLANTS OF  $F_1$  HYBRIDS OF *Hemerocallis Thunbergii*  $\times$  *H. aurantiaca* WERE CROSS-POLLINATED IN ALL POSSIBLE COMBINATIONS. THE RECIPROCALLS THAT GAVE + AND — REACTIONS ARE ENCLOSED IN CIRCLES

		Pollen Parents											
		5	3	1	9	2	6	12	4	10	11	7	8
Seed Parents	4	+	+	+	+	+	+	+	S	+	+	+	+
	9	+	+	+	F	+	+	+	+	+	+	—	+
	8	+	+	+	+	+	+	+	+	—	+	+	F
	2	+	+	+	+	F	—	+	+	+	+	+	+
	1	+	+	S	—	+	+	+	—	+	+	+	+
	6	—	+	+	+	+	F	+	+	+	—	+	+
	12	+	+	+	+	+	+	S	—	—	+	—	—
	10	+	+	+	+	+	+	+	—	S	—	—	—
	11	+	+	—	+	+	—	—	+	+	S	—	—
	7	+	—	+	+	—	+	—	—	—	—	S	—
	3	+	S	+	—	—	—	—	+	—	—	—	—
	5	S	—	+	—	—	—	—	—	—	—	—	—

Also very early in the investigations it was found that reciprocal crosses may give extremely different results (Stout, 1916). Thus a plant *A* may be highly compatible with the pollen of *B*, but *B* may be completely incompatible with pollen of *A* which is fully able to function in certain other cases. This condition has continually been found in all cultures studied whether the individuals were the progeny of selfings or of crossings. Sirks (1917) also obtained this result with *Verbascum phoenicium* throughout the earlier years of his studies.

Only recently has such + and — reactions for reciprocals been found in *Nicotiana* by East and Mangelsdorf (1925). They consider that this situa-

tion arises when self-incompatible plants are selfed by premature pollination. It is considered that the progeny of prematurely selfed  $S_1S_2$  plants will be composed of the Classes  $S_1S_1$ ,  $S_1S_2$  and  $S_2S_2$ . Then if premature pollinations are avoided the  $S_1S_1$  plants may be fertilized from  $S_2$  pollen of  $S_1S_2$  plants but the reciprocal will be fully incompatible. The plants giving different reciprocal reactions should fall into definite groups. In none of these plants that I have studied have the + and — reciprocals shown definite group reactions. The various points observed regarding the cross-relations may be illustrated by the behavior recorded in TABLES 2 and 3.

It is to be noted that this series of twelve plants are the  $F_1$  progeny of a cross between two different day lilies. All these plants are highly able to function both as male and female parents in fully compatible relations of fertilization. Four of the 12 were more or less self-compatible.

TABLE 3  
THE + AND — RECIPROCALLS COLLECTED FROM TABLE 2

	—AS POLLEN PARENTS										
	5	2	3	4		7	8	9	10	11	12
	3	2			6		8	9	10	11	12
	7	2		4			8				
	11	1					8				12
	12			4			8		10		
	10			4						11	
	1			4				9			
	9					7					
	2				6						
	4										
	6										
	8										

Of the 66 reciprocals possible for the 12 plants in question, 31 were + and —, and every plant was involved in such relations. Plants Nos. 4, 6 and 8 were — only, and No. 5 was + only. Such groups as could be made consist of a few plants and are not exclusive. Thus Nos. 3, 5 and 7 are + to pollen of Nos. 2 and 8, but of these three only 3 and 5 are + with 9, 10, 11 and 12, and only 3 is + to 6. A suggestion of a group reaction is seen only for the plants 3 and 5.

Turning to the groups of plants that are mutually cross-sterile (TABLE 2) they are as follows: 3 and 7; 5 and 6; 6 and 10; 6 and 11; 7 and 10; 7 and 11; 7 and 12, and 8 and 10. In no case does an intra-sterile group comprise more than two plants. It will also be noted that the groups are not exclusive. The groups of plants mutually cross-fertile are in larger numbers and are still less exclusive. For example, the intra-fertile groups involving plant No. 1 are 1 and 3; 1, 2 and 10; 1, 2 and 12; 1, 2, 6 and 8; 1, 6 and 7; and 1, 6 and 12.



In no particular is there a definite and consistent group relation. This irregular behavior of the day lilies as to cross-compatibility and cross-incompatibility is quite in agreement with that of the self-compatible and self-incompatible plants.

The assumed heredity factors for self- and cross-incompatibility, the  $S_1$ ,  $S_2$ , or  $S_3$  factors must be assumed to vary in values from 1 to 0 with decided lack of stability. When submitted to rigid tests of selection and inbreeding a stable value of either 1 or 0 is not maintained. Our most extended evidence of this is secured from continuous cultures of as many as seven generations. I know of no other study where this particular point has been so fully tested.

It is clear, therefore, that at least in the species studied by the writer the character of self-compatibility, provided this was the original condition of the species, is not necessarily eliminated by a sudden and discontinuous loss or mutation of definite units in heredity or by the acquisition of independent units with stable values. Possibly in time the genes involved may emerge from a condition of eversporting valuations to that of stability.

It seems to me that in these cases we shall do better to adopt as a working hypothesis the conception that the self- and cross-incompatibilities are determined by complex chemical compounds which are sufficiently labile to permit their passing readily through series of changes which are characterized by varying degrees of stability.

From our experience there is little hope of developing fully self-compatible races of plants in those species now highly self-incompatible. Among such species, especially when they are annuals, cross-pollination must be relied upon for the most complete yields of fruit from all the individuals. Yet among many such plants there is sufficient self-compatibility under selection to admit of line breeding if this be desired.

#### LITERATURE CITED

- Correns, C., 1912. Selbststerilität und Individualstoffe. Festsch. Med. Nat. Ges. Z. 84. Versam. Deutsch. Naturf. u. Ärzte.
- 1913. Selbststerilität und Individualstoffe. Biol. Centralb. 33: 389-423.
- East, E. M., and Mangelsdorf, A. J., 1925. A new interpretation of the hereditary behavior of self-sterile plants. Proc. Nat. Acad. Sci. 9: 166-171.
- Filzer, Paul, 1926. Die Selbststerilität von *Veronica syriaca*. Zeits. Indukt. Abstam. 41: 137-197.
- Lehmann, E., 1919. Ueber die Selbststerilität von *Veronica syriaca*. Zeits. Indukt. Abstam. 21: 13-40.
- 1922. Ueber die Selbststerilität von *Veronica syriaca*. II. Zeits. Indukt. Abstam. 27: 161-177.
- Sirks, M. J., 1917. Stérilité, auto-incompatibilité et différenciation sexuelle physiologique. Arch. Neerl. Soc. Exactes et Natur. Series III B. 3: 205-234.
- Stout, A. B., 1916. Self- and cross-pollinations in *Cichorium Intybus* with reference to sterility. Mem. N. Y. Bot. Gar. 6: 333-454.
- 1917. Fertility in *Cichorium Intybus*: The sporadic occurrence of self-fertile plants among the progeny of self-sterile plants. Amer. Jour. Bot. 4: 375-395.
- 1918. Fertility in *Cichorium Intybus*: Self-compatibility and self-incompatibility among the offspring of self-fertile lines of descent. Jour. Genetics 7: 71-103.
- 1920. Further experimental studies on self-incompatibility in hermaphrodite plants. Jour. Genetics 9: 85-129.
- 1922. Cyclic manifestation of sterility in *Brassica pekinensis* and *B. chinensis*. Bot. Gaz. 73: 110-132.





Plants of *Lilium Henryi* grown from seed obtained by crossing self-incompatible plants.

A. The most highly self-compatible plant thus far found in any species of *Lilium*. Every one of the seven flowers selfed (see the round tags) yielded good capsules. The two capsules marked *x* are from cross-pollinations with other plants of the same species. Such self-compatible plants occur sporadically among the progeny of parents fully self-incompatible.

B. A fully self-incompatible plant. No capsules resulted to the eight flowers selfed. The four good capsules (*x*) are from cross-pollination and prove that the plant is able to function as a seed parent. Three flowers (*x*) crossed with pollen of another species (*L. speciosum*) failed completely to form fruits, illustrating the lack of affinity between species.

These two plants exhibit extreme cases of self-compatibility and self-incompatibility as they regularly occur in many species. Neither of these conditions breed true. In the progeny of A there will be many plants fully self-incompatible. Among the progeny obtained by crossing self-incompatible plants some that are more or less self-compatible will be found.





Typical cases of self-compatibility and self-incompatibility in Chinese cabbage.

1. Highly self-compatible plant with highest degree of self-fruitlet during mid-period of bloom.

2. Plant with "medium" self-compatibility beginning early in the period of bloom.

3. Plant medium self-compatible rather late in the period of bloom.

4 and 5. Plants completely self-incompatible. The fruitlets seen on 5, 4 and at tip of 3 are from cross-pollination.

The extreme grades of self-compatibility and self-incompatibility here shown continue to appear among progenies even where there is repeated selection of parents that are as highly self-compatible as No. 1.





## INHERITED POLLEN-STERILITY IN SHEPHERD'S-PURSE

GEORGE H. SHULL

*Princeton University*

(WITH PLATE 30)

From among six—perhaps seven—fundamentally different kinds of sterility which I have encountered in my genetical studies with shepherd's-purse (*Bursa bursa-pastoris* and allied species), I have chosen for presentation to this Conference an example of fertility and sterility which is conditioned by the presence of one or the other factor of a single Mendelian pair. I have assumed that this particular kind of sterility would be perhaps less likely than some other kinds to be contributed to this program by other workers, though pollen-sterility has been noted in many other plants, and in at least one, the sweet pea, it has been studied genetically (Bateson, Saunders and Punnett, 1905, pp. 91-92) and shown to be in that case a Mendelian recessive just as I find it to be in shepherd's-purse.

### THE TRAIT

The particular type of sterility which is here presented consists in the complete or nearly complete abortion of the pollen, producing shrunken or "contabescent" anthers, so named by Gaertner (1844) who gave the first discussion of this type of sterility, though it had also been noted before him by Koelreuter. "Contabescence" is also briefly discussed by Darwin (1876, 2: 149-150):

While no cytological studies have been made on the pollen-sterile plants in my cultures, anatomical studies have shown that the anthers of pollen-sterile and pollen-fertile plants can not be distinguished from each other up to the pollen mother cell stage, after which there is rapid disintegration of the tissues within the loculi of the contabescent anthers, leaving them, at the period of anthesis, nearly or quite empty (see PLATE 30). It is very probable, therefore, that cytological studies on these pollen-sterile Bursas will show them to be in essential agreement with the pollen-sterile *Lathyrus* discovered by Bateson, Saunders and Punnett, and studied cytologically by Gregory. Gregory (1904, p. 91) found that in the contabescent anthers of *Lathyrus* the cell divisions proceed normally up to the time of meiosis, at which time "the longitudinal fission of the spireme takes place quite normally, but the segmentation into chromosomes is, if carried out at all, irregular, and the pollen mother cells degenerate."

In external appearance the anthers are slightly smaller than in the pollen-fertile plants, the sides are rather sunken and there is usually no indication or only slight indication of a dehiscence of the loculi. The plants take on a characteristic appearance, because they can not be self-fertilized,



FIG. 1. A pollen-fertile, pollen-sterile chimaera in family 24449. The right-hand branch was fully pollen-fertile, while the rest of the plant was pollen-sterile.

and the capsules, therefore, fail to develop unless pollen from some other source is applied.

FIGURE 1 shows an interesting chimaera from family 24449 in which one large branch of the plant had perfect flowers (pollen-fertile), while the other branches had flowers with contabescent anthers. That the pistils and their included ovules containing the eggs, are not correspondingly deranged is easily demonstrated by applying pollen from some pollen-fertile plant, and the same thing is also shown naturally when the plants are left unguarded, for the visit of an insect carrying *Bursa* pollen is recorded on the pollen-sterile plant by the formation of a group of capsules well-filled with seeds, thus giving the same general aspect as in the self-sterile species, *Bursa grandiflora*, where likewise capsules do not develop except as records of the advent of foreign pollen. The pollen-sterile portion of the chimaera shown in FIGURE 1 was successfully fertilized by the pollen from the perfect-flowered portion of the same plant, but only six offspring were secured (family 25424), one fully fertile and five pollen-sterile. While this is a poor fit for a 1:1 ratio, it is at least clear that the fertile branch of the chimaera was heterozygous, and it is probable that the pollen-sterile sector was genotypically as well as phenotypically of the recessive type.

#### ORIGIN OF POLLEN-STERILITY

This peculiar form of sterility was first observed in *Bursa* in 1917 in three  $F_2$  hybrid families produced by pollinating a specimen of *Bursa orientalis* (Shull ined.) from Singen, Korea, and a specimen of *B. Heegeri* (from family 14347) which had segregated from previous crosses with *B. bursa-pastoris*. These three families showed, respectively:

FAMILY	POLLEN-FERTILE	POLLEN-STERILE
16570	202	83
16571	202	61
16572	150	69
Total	554	213
Ratio	2.6:1	

The same character appeared independently the next year (1918) in an  $F_2$  family (17611) from a cross between *Bursa bursa-pastoris* received from Saharanpur, India, with *B. Heegeri* (from family 15358). The segregation of this family resulted in 156 pollen-fertile to 77 pollen-sterile, a ratio of almost exactly 2:1. Again the following year (1919) this particular type of sterility presented itself, again independently, in nine  $F_2$  families produced by crossing *B. bursa-pastoris* from four collections made at Shanghai, China, with two different specimens of *B. Heegeri* (seven of the families from crosses with a *Heegeri* from family 16304, and two with a *Heegeri* from family 16610). The total of these nine families consisted of 371 pollen-fertile to 143 pollen-sterile, a ratio of 2.6:1 (TABLE 1). It also appeared the same year in an  $F_2$  (18319) between a *B. bursa-pastoris* from

Stockholm, Sweden, and *B. Heegeri* (from family 16545), in which there appeared a split into 234 pollen-fertile to 76 pollen-sterile, a ratio of 3.08:1.

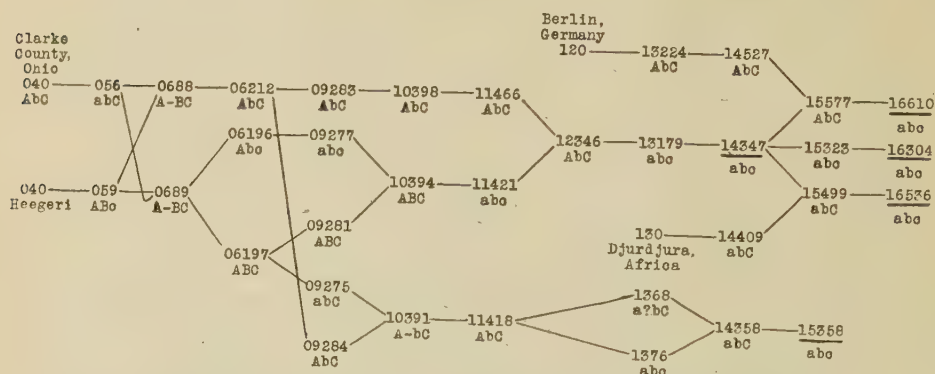


FIG. 2. Pedigree of the five *Bursa Heegeri* plants (heavily underscored) which were used as pollinators in the crosses in the  $F_2$  of which pollen-sterility appeared as recessive segregates. The numbers are family numbers and the literal symbols under each number indicate the rosette and capsule form of the individual which served as the parent of the next succeeding family: AB = *heteris*, Ab = *tenuis*, aB = *rhomboidea*, ab = *simplex*, C = triangular capsules as of *B. bursa-pastoris*, c = round capsules as of *B. Heegeri*. In the case of crosses, the maternal family number is always given above, the paternal number below. All numbers connected with preceding numbers by a single line represent families which resulted from guarded self-fertilizations. The extreme left-hand number of each line originated from wild or uncontrolled material.

TABLE 1

SEGREGATIONS OF POLLEN-STERILITY IN  $F_2$  FAMILIES DERIVED FROM THREE CROSSES BETWEEN *B. bursa-pastoris* FROM SHANGHAI, CHINA, AND *B. Heegeri*

FAMILY NUMBERS					POLLEN-FERTILE	POLLEN-STERILE
♀	P <sub>1</sub>	♂	F <sub>1</sub>	F <sub>2</sub>		
16638		16304	17328	18426	43	16
16639		16304	17329	18427	24	5
16639		16304	17329	18428	37	10
16640		16304	17330	18429	30	17
16641		16304	17331	18430	184	66
16641		16304	17331	18431	7	4
16641		16304	17331	18433	4	2
16641		16610	17332	18434	7	3
16641		16610	17332	18435	35	20
Total.....					371	143

While these four origins of pollen-sterility were quite independent of each other so far as any intentional study of the subject was concerned, it will have been noted that each apparently independent origin of this trait took place in an  $F_2$  family of a cross in which *B. Heegeri* had served as the pollen parent. On this account it seems more simple to suppose that pollen-sterility was brought into these crosses from *B. Heegeri* than that the same trait should have originated independently in wild material from four dif-



ferent localities so widely separated as Singen, Korea; Shanghai, China; Saharanpur, India; and Uppsala, Sweden. A similar, but not quite as formidable, difficulty is presented for the supposition that all of these unexpected origins of pollen-sterility involved a single origin in the strain of *B. Heegeri* which was being used as pollinators, for the same individual *Heegeri* was not used for any two of these crosses, except that one plant of 16304 served as pollinator for four of the five crosses in the Shanghai material. The relationship of these *Heegeri* pollinators to one another is shown in FIGURE 2. It is seen that four of these were closely related to each other, while 16358 was connected with the rest only in the seventh preceding generation. It must be considered also that in the course of twelve years preceding the first discovery of pollen-sterility, I had made over a hundred distinct crosses in which *B. Heegeri* had served as the pollinators, and that in that same year (1917) I had F<sub>2</sub> families derived from 51 such crosses, which might have exhibited a segregation with respect to pollen-sterility. It is not unlikely that pollen-sterility did actually occur in some of these other families, but was not noted because it was obscured by sterility induced by unfavorable environmental conditions. It is clear, at any rate, that in no other family did such segregation occur with sufficient distinctness to attract attention, even when the cases had been noted in the families derived from the Korea material. Although it seems likely that pollen-sterility came in through the *Heegeri* pollinators, it is not possible to get conclusive proof of this and we must, therefore, leave the origin of pollen-sterility in shepherd's-purse, as origins must be so often left, in the realm of partial obscurity.

#### POLLEN-STERILITY IN SUBSEQUENT GENERATIONS

While the inheritance of this trait interested me intensely when first noted, it was allowed to remain secondary to my interest in other problems with which I was fully occupied, until 1921, when I began to actively study the matter. During the interval between the first record of pollen-sterility, and this taking up of active investigation of pollen-sterility as such, the trait had been observed and recorded in 32 families in addition to the 14 original families already presented. These 32 families in which pollen-sterility was incidentally recorded are assembled in TABLE 2. The data in this table are of unequal value, as records did not always include whole families, and in some cases no account was taken of the likelihood that non-hereditary sterility was present in the same family. They are given here for the sake of completeness, and also because they suggest in certain cases relationships which have not yet been investigated. It is seen that in 15 of these 32 families the ratios were conspicuously near the monohybrid ratio; 11 showed a marked excess of sterile individuals, probably through the incidence of a wholly different kind of sterility, for these 11 families were so located that their illumination was materially reduced, and much sterility, presumably wholly non-hereditary, is usually observed when shepherd's-

purse is grown in poor conditions with respect to illumination. Of the remaining 6 families one contained only 5 individuals, 3 pollen-fertile and 2 pollen-sterile, and might belong to any type of segregation, while 5 showed such low incidence of pollen-sterile plants as to suggest the possible occurrence of duplication of the fertility factor. Of one of these families (18310) the record is indefinite, but only "several" pollen-sterile plants were observed in a family of 92 individuals which was recorded as "mostly quite fertile"; the others (18306, 19333, 19554, 20540) gave ratios ranging from 7:1 to 19:1.

### THE RESULTS OF DIRECT EXPERIMENTAL ANALYSIS

In 1921 I began some specific experiments on the inheritance of pollen-sterility by selfing several fertile plants in family 20406 (see TABLE 2).

TABLE 2  
FAMILIES FROM SEVERAL DIFFERENT ORIGINS, IN WHICH POLLEN-STERILITY WAS INCIDENTALLY RECORDED

FAMILY NUMBERS		POLLEN-FERTILE	POLLEN-STERILE
F <sub>1</sub>	F <sub>2</sub>		
16572	17360	13	11
	17361	49	28
	17363	28	32
	17364	69	35
	17365	31	70
	17366	57	32
	17367	45	31
	17368	46	47
	17369	41	53
	17452	162	28
16571	17453	93	76
	17455	210	91
17314	18282	15	5
17328	18304	226	61
	18305	113	38
17329	18306	69	3
	18307	3	2
17330	18308	20	6
	18309	179	62
17331	18310	—*	—*
17332	18313	47	19
	18314	11	6
	18315	137	68
17452	18316	244	84
17351	18363	227	91
18363	18532	72	12
18365	19333	59	8
19332	19554	97	8
	20404	14	3
19333	20405	235†	87
19554	20406	100	24
	20540	94	5

From these selfings were grown in 1922 three families, one of which (21494) consisted solely of 308 pollen-fertile plants, while the others (21492 and 21493) split respectively into 152 pollen-fertile to 51 pollen-sterile, and

\* Several were sterile, but mostly quite fertile.

† Thirty-one of these were described as largely sterile.

206 pollen-fertile to 102 pollen-sterile. The close approximation to a 2:1 ratio in the last of these families led me to make a considerable number of self-pollinations in this family in order to test the ratio between heterozygotes and homozygotes. The results are assembled in TABLE 3, but are somewhat indecisive because I had not yet fully appreciated the importance of taking precautions against the occurrence of non-hereditary sterility induced by unfavorable environmental conditions, especially inadequate illumination.

TABLE 3

SEGREGATIONS FROM SELF-FERTILIZED PLANTS IN FAMILY 21493, WHICH WAS THE PROGENY OF A SELF-FERTILIZED PLANT IN FAMILY 20406 (TABLE 2)

FAMILY NUMBERS	POLLEN-FERTILE	POLLEN-STERILE
22369	20 (?)	0
22375	112	0
22372	118	7
22374	112	7
22376	87	4
22367	18	2
22368	15	5
22370	17	3
22371	75	49
22373	12	7
22377	42	14
22378	15	2
22379	98	27
22380	23	11
22381	49	15

Since the parental family had exhibited a monohybrid composition, there was no theoretical grounds for expecting the occurrence of dihybrid ratios in any of these families, but reference to TABLE 3 shows that three families closely approximated the 15:1 ratio, while several others deviated rather widely from 3:1; but, owing to the small numbers in most of these other families no significance could be attached to the ratios in them. Only two of the fifteen families contained no pollen-sterile individuals, but if the small number of sterile individuals in the three families which approximated the 15:1 ratio owed their sterility to inadequate illumination or some other unfavorable condition, these three families must be classed as the offspring of homozygous pollen-fertile parents, thus changing the grouping of the families to 5 non-splitting and 10 splitting, just as would be expected in a monohybrid segregation.

The same type of experiment was continued in a small way the next year by selfing seven individuals in family 22380 (TABLE 3) which had again shown a close approximation to a 2:1 ratio. Three of these seven lots of seeds gave no germinations, but four families were grown from these selfings in 1924 with the results presented in TABLE 4.

It is seen that here again the segregations rather closely approximate a monohybrid ratio, but in every case with a slight excess of pollen-sterile plants above the number expected.

TABLE 4  
SEGREGATIONS OF POLLEN-STERILITY IN PROGENIES FROM SELF-FERTILIZED POLLEN-FERTILE  
PLANTS IN THE MONOHYBRID FAMILY 22380 (TABLE 3)

FAMILY NUMBERS	POLLEN-FERTILE	POLLEN-STERILE
23340	232	86
23341	209	85
23342	229	88
23343	237	83
Total	907	342
Expected (3:1)	936.75	312.25

At the same time that the selfings were made in family 21493 which gave the families listed in TABLE 3, I also fertilized three pollen-sterile plants in the same family with pollen from as many of their pollen-fertile sibs. These crosses gave rise to three families having the following composition:

FAMILY NUMBERS	POLLEN-FERTILE	POLLEN-STERILE
22382	25	18
22383	63	14
22384	58	19

While the first of these families can easily be assumed to represent a 1:1 segregation, the other two show a close approximation to 3:1 and these, even more strikingly than the 15:1 ratios of TABLE 3, suggest the occurrence of a duplication of the fertility factor. The question of duplication of this factor has not yet been adequately investigated, however, and this question must be left entirely on one side for a later report. I can only say that present evidence suggests that the fertility factor is sometimes duplicated, but that such duplication is probably relatively rare. A considerable number of new outcrosses of the pollen-sterile individuals have since been made, in a special attack on this phase of the problem.

As the number of individuals in the  $F_1$  family (22382) between a pollen-sterile plant and one of its pollen-fertile sibs, which had indicated a monohybrid ratio, was rather small and the deviation from the expected 1:1 ratio somewhat marked, I decided to test the reality of its monohybrid nature by a breeding test. Four of the fertile individuals in this family, self-fertilized, gave in 1924 the results shown in TABLE 5.

The moderate excess of the pollen-sterile class may again indicate the

TABLE 5  
SEGREGATIONS OF POLLEN-STERILITY FOLLOWING SELF-FERTILIZATION OF HETEROZYGOTES IN  
FAMILY 22382

FAMILY NUMBERS	POLLEN-FERTILE	POLLEN-STERILE
23340	232	86
23341	209	85
23342	229	88
23343	237	83
Total	907	342
Expected (3:1)	936.75	312.25



addition of a few cases of induced sterility, and we may consider the results a satisfactory confirmation of the assumption that the grandparent was a monohybrid with respect to a pollen-fertility factor.

Because both the self-fertilizations and the sib-crosses in family 21493 suggested the possible presence of both monogenic and digenic pollen-fertility and left in some doubt the relative ratio of homozygotes and heterozygotes, it seemed desirable to test further the segregations of this factor in what seemed to be monohybrid splitting families. The families given in TABLE 5 were so consistent that it was decided to repeat in these families the tests for homozygotes and heterozygotes. A large number of pollen-fertile plants in the four families of TABLE 5 were selfed, and 29 families were grown during the past winter (1925-26) from these selfings. The results are given in TABLE 6. Ten of these families, containing a total of 1,121 plants, showed one (24450) that had one apparently sterile plant to 124 fertile, one (24437)

TABLE 6

SEGREGATIONS OF POLLEN-STERILITY FROM SELF-FERTILIZED FERTILE PLANTS IN THE MONOHYBRID FAMILIES 23340, 23341 AND 23342 OF TABLE 5

FAMILY NUMBERS		POLLEN-FERTILE	POLLEN-STERILE
F <sub>1</sub>	F <sub>2</sub>		
23340	24436	122	—
	24438	117	—
	24444	125	—
	24454	124	—
	24455	123	—
23341	24456	125	—
	24459	123	—
23340	24437	66	3
	24441	60	8
	24450	124	1
	24435	94	31
	24439	95	30
	24440	99	24
	24442	91	33
	24443	94	31
	24445	92	32
	24446	93	26
	24447	98	20
	24449	100*	22
	24451	94	31
	24452	99	25
	24453†	89	36
	24457†	87	38
	24458	94	29
23341	24460	86	32
	24461	96	27
	24462	90	33
	24463	84	41
23342	24464	75	50

\* The fertile-sterile chimaera illustrated in figure 7 was included in this group.

† These grew in the coldframe where the illumination was less adequate than in the greenhouse, and less evenly distributed.

had 3 sterile plants to 66 fertile, and one (24441) had 8 sterile plants to 60 fertile, while the remaining 19 families showed close approximation to the monohybrid ratio, except that families 24453, 24457, 24463 and 24464 showed a marked excess of sterile plants, probably due to induced sterility. These last four families contained 335 fertile to 165 sterile, or almost exactly 2:1 while the remaining 15 families contained 1415 fertile to 426, the expectation on the basis of a 3:1 ratio being 1380.75 to 460.25.

If we allow the several families with abnormally low incidence of sterility to stand as non-segregating families in which a small amount of non-hereditary sterility has been induced by environmental conditions, the non-segregating and segregating families stand in the ratio 10:19, or as close as possible to the expected 1:2 ratio. The only doubtful points relate to the real character of the small number of families which deviated markedly from the 3:1 ratios, and the still smaller number of families which have so small a percentage of pollen-sterile plants as to suggest a duplication of factors. The 2:1 ratios have already been shown to yield segregations chiefly in 3:1 ratios, and the regular occurrence of these monohybrid ratios makes improbable the occurrence of true dihybrid ratios in the same families. This question is being actively investigated and will be made the subject of a later report.

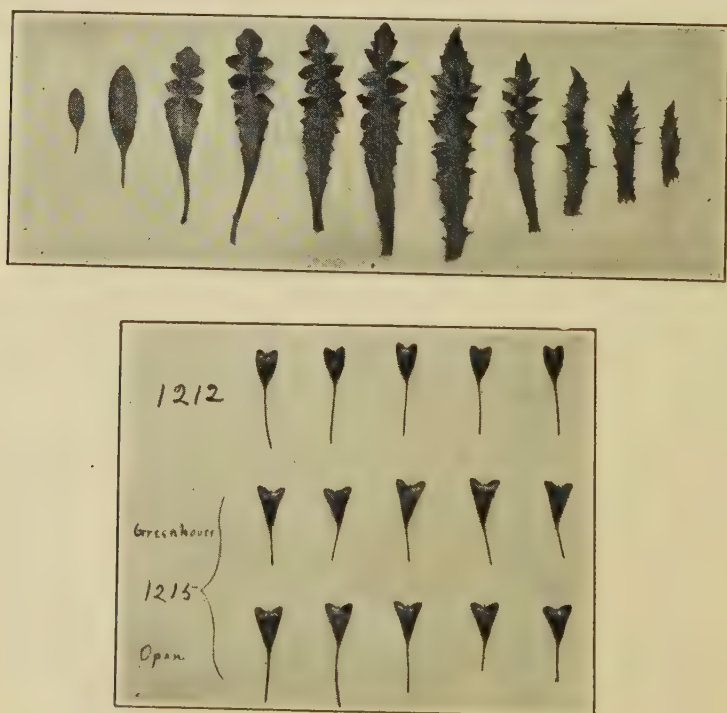


FIG. 3. Climax leaves of the rosettes in *Bursa penarthae* showing the rather low-grade *rhomboidea* lobing. The capsules of this species (1215) are also given, with those of *B. bursa-pastoris* (1212) from the same region, for comparison. The pedigree numbers were afterward changed to 12360 and 12357, respectively.

# LINKAGE OF THE POLLEN-STERILITY FACTOR WITH A LEAF-LOBE FACTOR

There remains to be considered a remarkable relationship between the pollen-sterility factor and a leaf-lobe factor, which probably represents the first case of linkage observed in shepherd's-purse. It was noted first in 1924 in four families, produced by selfing four *subrhomboidea* plants in family 22405, that there was a simultaneous segregation both with respect to leaf-lobing—into *subrhomboidea* and *simplex*—and with respect to fertility—into pollen-fertile and pollen-sterile—the distribution not being independent, but so correlated that, in the main, only the *subrhomboidea* class split into pollen-fertile and pollen-sterile, while the group characterized by *simplex* rosettes rarely showed pollen-sterility. The families illustrating this relationship are presented in TABLE 7.

TABLE 7

SIMULTANEOUS SEGREGATION WITH RESPECT TO POLLEN-STERILITY AND *subrhomboidea* LEAF LOBING, SHOWING THE FIRST CLEAR CASE OF LINKAGE IN SHEPHERD'S-PURSE

FAMILY NUMBERS			SUBRHOMBOIDEA			SIMPLEX		
P <sub>1</sub>	F <sub>1</sub>	F <sub>2</sub>	FERTILE	STERILE	FERTILITY NOT NOTED	FERTILE	STERILE	FERTILITY NOT NOTED
21331	22405	23316	65	33	..	25	1	..
		23317	46	24	..	39	0	..
		23318	54	25	..	45	0	..
		23333	73	7	..	42	3	..
		24419	63	22	..	34	0	..
		24420	60	29	..	26	0	..
	23317	24421	57	22	6	31	1	2
		24422	70	26	1	32	1	..
		24423	62	17	4	36	0	..
		24425	54	23	4	42	0	..
		24426	56	29	..	39	1	..
		24427	57	17	..	49	0	..
22405	23318	21428	50	12	..	56	4	..
		24429	59	19	..	46	1	..
		24431	46	15	..	60	1	..
		24432	55	24	..	42	1	..
		24433	60	23	..	39	1	..

Three families from the same series of self-pollinations are not included in the table because of incompleteness of the records. Of these excluded families, 24418 and 24430 were only partially recorded with respect to the leaf character; with respect to pollen-sterility these segregated into:

(24418) 84 pollen-fertile, 33 pollen-sterile;

(24430) 76 pollen-fertile, 44 pollen-sterile.

In the case of family 24418, a number of rosettes were still capable of determination when the classification was made with respect to pollen-sterility, and it was noted that all of the pollen-sterile plants which still exhibited their rosette character had *subrhomboidea* rosettes. In family 24424, likewise from the same set of breedings, the rosettes were classified but not the pollen-sterility, the segregation showing 57 *subrhomboidea* and 38 *simplex*.

THE *SUBRHOMBOIDEA* CHARACTER AND ITS ORIGIN

On September 23, 1913, Professor A. H. Trow collected for me at Penarth, near Cardiff, Wales, seeds of a form of shepherd's-purse which, from its distinctive characteristics and breeding behavior, has been considered a new species and named *Bursa penarthae* (Shull ined.). The relation of this form to the families presented in TABLE 7 is indicated in FIGURE 4. The rosette leaves of *B. penarthae* were of a peculiar modified form of *rhomboida* in which there was a partial suppression of the deep lobing usual to that rosette type (FIG. 3).

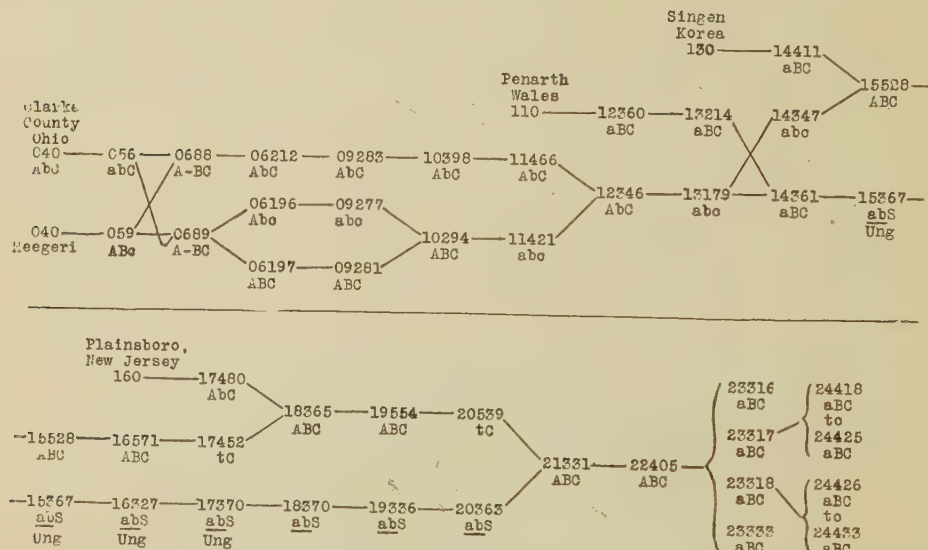


FIG. 4. Pedigree of the families included in Table 7 which exhibited apparent linkage between *subrhomboida* leaves and pollen-sterility. Literal symbols have the significance indicated in Fig. 2, with the addition that *abS* indicates *simplissima*, *t* = *tenuiloba*, and that all breeding was controlled as indicated by the lines, except that those marked "Ung" were open-pollinated. The breeding results show that each of these was in fact self-fertilized though not artificially controlled.

A cross of *B. penarthae rhomboida*, with *B. Heegeri simplex* resulted in an  $F_1$  of rather low-grade *rhomboida* displaying much sterility, which segregated in the  $F_2$ , not only the expected *rhomboida* and *simplex* types but also a wholly new type of rosette in which the leaves were nearly or quite unlobed (FIG. 5), which has since been designated "*simplissima*."

A strain of this *simplissima* segregate (symbolized by *abS* in Fig. 4) has been maintained and used in a number of experiments which have shown repeatedly that *simplissima* is a *rhomboida* plus a recessive inhibitor. The original *simplissima* plants in family 15367 were rather weak-stemmed and not very fertile, so that for several years this strain was continued by the use of unguarded seeds—a procedure which could be followed without serious detriment, because of the recessive nature of the *simplissima* inhibitor.

In 1921 a plant of this *simplissima* strain (family 20363) was used to pollinate a castrated specimen of *tenuiloba* from family 20539. (For the



origin, description and figures of *tenuiloba* see Shull, 1927). The  $F_1$  family (21331) became somewhat diseased and characterization was uncertain, but it was clear that the plants were not exhibiting the peculiarities of either parent, but were more like the ordinary forms of *heteris* and *rhomboidea*. One plant of each of these two rosette types was self-fertilized and the  $F_2$  families were grown in 1922. The apparently *rhomboidca*  $F_1$  plant gave a progeny (family 22406) of 45 offspring which included *tenuiloba*, *heteris*, and *tenuis*

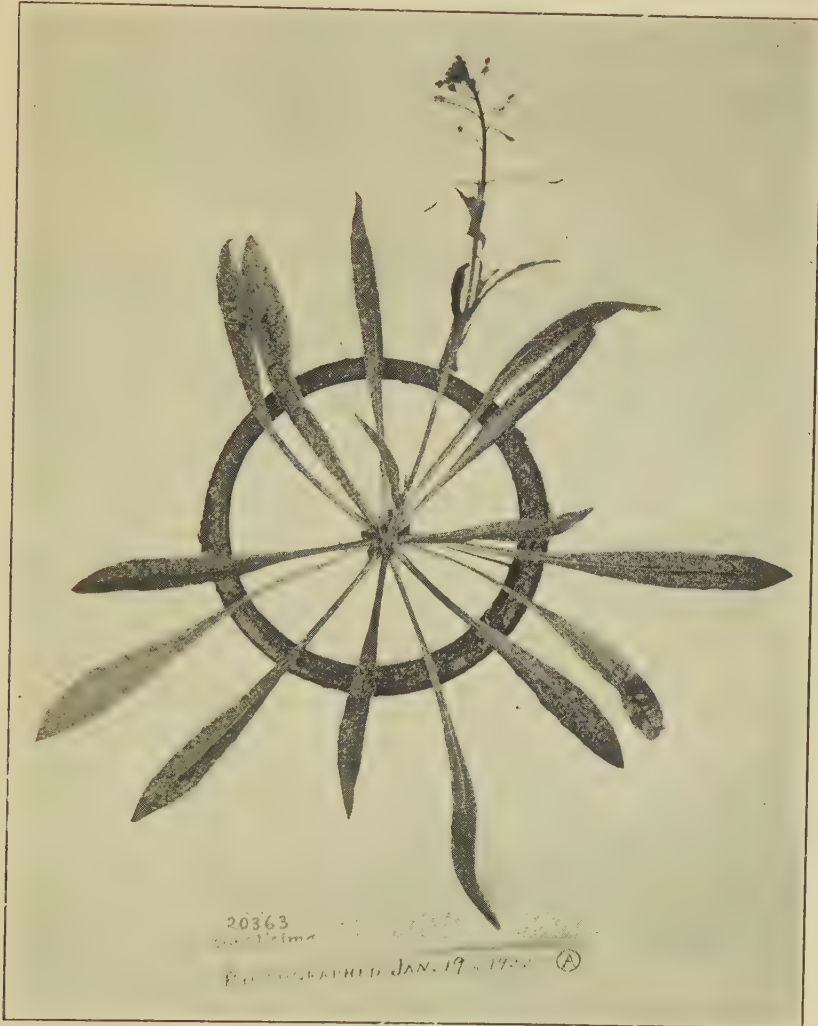


FIG. 5. A *simplissima* rosette derived from a cross between *B. penarthae rhomboidea* and *B. Heegeri simplex*.

specimens, thus showing that the  $F_1$  "*rhomboidea*" mother was genotypically a *heteris*. The  $F_1$  *heteris* plant gave an  $F_2$  progeny (family 22405) of 289 plants which included the most complex array of forms which has yet been

exhibited in any of my *Bursa* families, containing plants of *heteris*, *rhomboidea*, *subrhomboidea*, *tenuis*, *simplex*, *heteroloba*, *tenuiloba*, *revoluta*, *serulata*, and a whole heterogeneous group of small, smooth, bluish-green plants to which the name "*cyanea*" was given. This motley array of forms has not yet been fully analyzed, so that their relations to one another cannot be completely stated. The "*subrhomboidea*" plants were considered merely as low-grade *rhomboidea* whose sinuses hardly reached the midrib. One of the supposedly *simplex* plants, self-fertilized, produced family 23333 in 1924, consisting of 80 *subrhomboidea* and 45 *simplex*, thus proving that the parent of this family was only apparently a *simplex*, but genotypically a *subrhomboidea*.

Just what the relationship is between *subrhomboidea* and *rhomboidea* has not yet been determined. It may result from a modified *rhomboidea* gene or from the presence of one or more separate modifying genes acting with the *rhomboidea* gene *B* to produce a low-grade lobing of the *rhomboidea* type.



FIG. 6. At left four climax leaves from rosettes of *subrhomboidea* and at right four of *simplex*, in the progeny (24425) from a self-fertilized *subrhomboidea*.

#### IS SUBRHOMBOIDEA LETHAL?

The low grade of lobing of the *subrhomboidea* form has made its separation from *simplex* a little insecure, but on the whole it is probable that the percentage of error in the classification has been low. Single leaves from each of several *subrhomboidea* and *simplex* segregates in one of the segregating families are illustrated in FIG. 6. From this figure it will be readily seen how slight modification would be required to make either class indistinguishable from the other. The relatively low incidence of *subrhomboidea* in three of the first four families of TABLE 7 suggested two possible explanations, namely, either that the difficulties of classification threw a considerable number of *subrhomboidea* plants into the *simplex* class, or that the homozygous *subrhomboidea* is lethal. This question is now under investigation. In favor of the first explanation is the fact that, as already stated, one such error has already been found, the *subrhomboidea* parent of family 23333 having been classed as a *simplex*. Against this explanation of deficiency of the *subrhom-*

*boidea* class is the relatively clear-cut separation of the pollen-sterile plants into the recognizably *subrhomboidea* class. The suppression of lobing and the accentuation of sterility in shepherd's-purse are both sometimes due to unfavorable environmental conditions, and it would be reasonable to expect, therefore, that there would be a higher instead of a lower percentage of sterile plants in the *simplex* class, if the defective ratios resulted merely from errors of classification. The absence of subjective bias in making the classification is assured by the fact that the grouping with respect to rosette characters and that with respect to fertility and sterility is separated by a considerable interval of time.

Favorable to the second explanation of the deficiency of the *subrhomboidea* class, namely, that homozygous *subrhomboidea* is lethal, are not only the 2:1 ratios, frequently observed, but also the fact that 14 self-fertilizations of the *subrhomboidea* segregates in families 23317 and 23318 produced not a single non-splitting progeny (see TABLE 7). Whether a similar result will be found in those families in which the *subrhomboidea* is not deficient remains to be determined. It may very well be that *subrhomboidea* is not itself lethal, but merely closely linked with a recessive lethal factor.

Inspection of the ratios in TABLE 7 brings to light an interesting series whose significance is not at present apparent. Without making any assumptions as to what ratios are theoretically expected, these ratios may be roughly classified as follows:

NUMBER OF FAMILIES	RATIO
5	2:1:1
4	2:1:2
4	3:1:2
1	3:1:3
1	3:1:4
1	4:1:5
1	10:1:6

Yet another question which requires testing is the status of the pollen-sterile specimens in the *simplex* class. Here again there are two suggestions: The most interesting possibility is that they are crossovers, produced by a break between the sterility factor and the *subrhomboidea* factor. Caution must be exercised, however, in interpreting these pollen-sterile *simplex* as crossovers, in view of the fact that *subrhomboidea* plants are occasionally indistinguishable from *simplex*. Until breeding tests have made certain that the pollen-sterile plants in the *simplex* group are genotypically *simplex*, the possibility that they are *subrhomboidea* which have been erroneously referred to *simplex* must remain a plausible interpretation.\*

\*Since this was written, two  $F_1$  families (25372, 25373), produced by crosses of pollen-sterile *simplex* in family 24432 (see Table 7) and a *B. Heegeri simplex*, have been grown. These have been uniformly *simplex*, thus proving that the mother was a true *simplex* and not a *subrhomboidea* which had been mistakenly classified as a *simplex*. It still remains possible that this pollen-sterile *simplex* plant in family 24432 owed its sterility to some other cause than the presence of the recessive Mendelian factor. Another generation must be grown before it can have been definitely demonstrated that the pollen-sterile *simplex* is produced by crossing over.



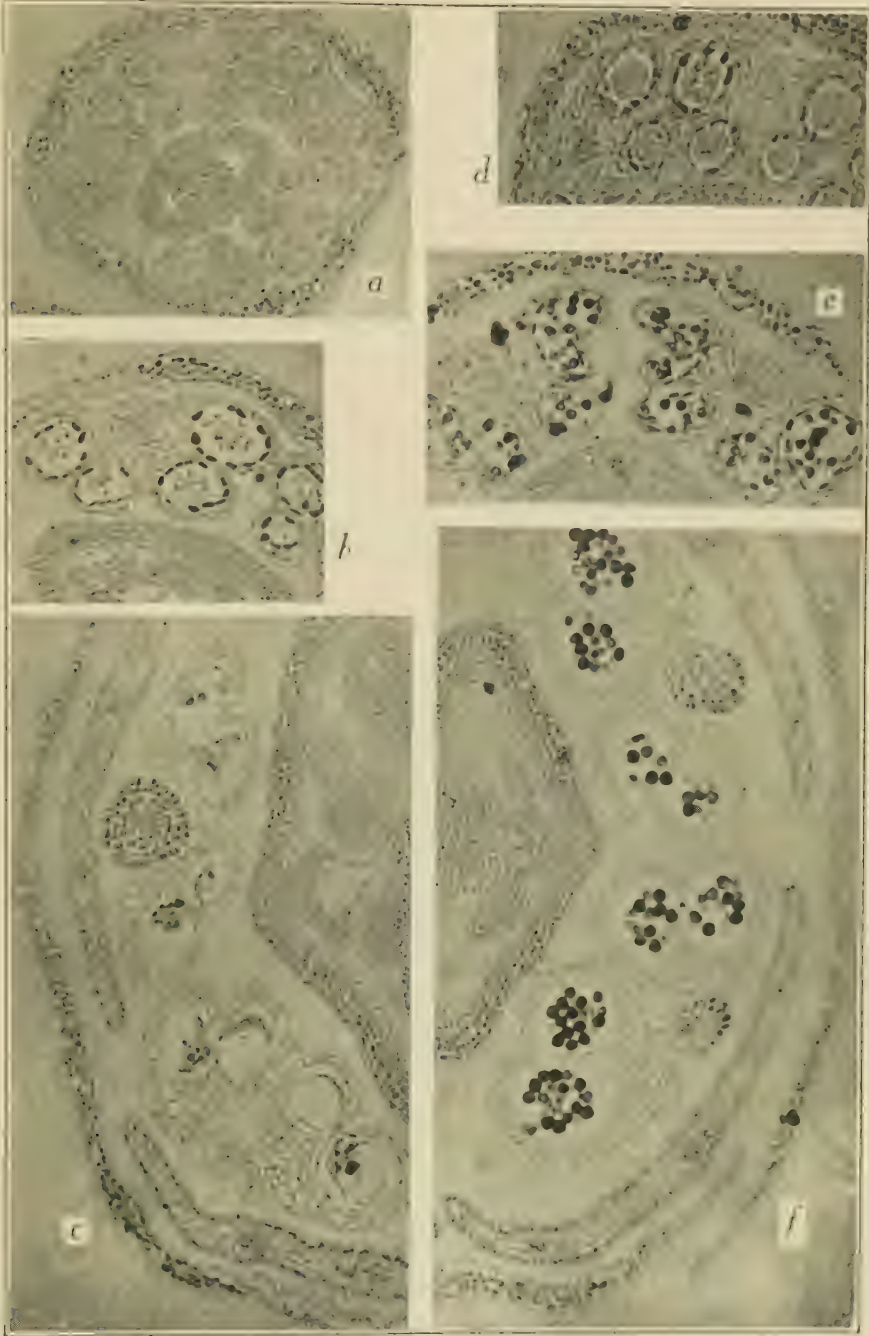
## SUMMARY

1. Pollen-sterility was observed in shepherd's-purse in 1917 to 1919 in ratios suggesting that it is a Mendelian recessive to pollen-fertility.
2. Subsequent breeding has confirmed this interpretation, but the results are often obscured or distorted by the simultaneous occurrence of another type of sterility which is induced by unfavorable conditions of the environment.
3. There are some indications that the factor for pollen-sterility is sometimes duplicated, but this suggested possibility requires further investigation.
4. In one series of families the pollen-sterility factor appears to be closely linked with a factor which produces a low-grade *rhomboidea*-like lobing of the leaves, which has been designated, "*subrhomboidea*."
5. The origin of *subrhomboidea* is described and it is noted that occasionally this form is not distinguishable from the *simplex* plants segregating from the same selfed parent. The breeding test is then necessary for its certain recognition.
6. The relationship between *subrhomboidea* and pollen-sterility is such that when both are segregating simultaneously, only the *subrhomboidea* group usually splits into pollen-fertile and pollen-sterile classes, while the *simplex* group shows pollen-sterility only rarely if at all.
7. Evidence is presented that *subrhomboidea* is either itself lethal in the homozygous state, or is closely linked with a recessive lethal.
8. It is not yet certainly demonstrated that the pollen-sterile plants in the *simplex* class are the result of crossing over between the *subrhomboidea* factor, *B<sub>r</sub>*, and the pollen-sterility factor *S<sub>p</sub>*. They may have resulted from errors of classification with respect to the leaf lobe character.

## LITERATURE CITED

- Bateson, W., Saunders, E. R., and Punnett, R. C., 1905. Experimental studies in the physiology of heredity. Report II to the Evolution Committee of the Royal Society (London), 154 pp. Sweet pea (*Lathyrus odoratus*), pp. 80-99.
- Darwin, C., 1876. The variation of animals and plants under domestication. 2nd ed., 2 vols. New York: D. Appleton & Co.
- Gaertner, C. F., 1844. Beiträge zur Kenntniss der Befruchtung der vollkommenen Gewächse. I. Versuche und Beobachtungen über die Befruchtungsorgane der vollkommeneren Gewächse, und über die natürliche und künstliche Befruchtung durch den eigenen Pollen. xii + 644 pp. Stuttgart: E. Schweizerbartsche Verlagshandlung.
- Gregory, R. P., 1904. The reduction division in ferns. Proc. Roy. Soc. London 73: 86-92.
- Shull, G. H., 1927. A heterozygous phenotype in shepherd's-purse. Hereditas 9: 225-235.





Sections of buds of pollen-fertile and pollen-sterile plants of shepherd's-purse. The microscopic slides from which these photomicrographs were made were prepared by Mrs. William W. Bennett. The magnifications of comparable stages in the two types are approximately the same, but are considerably higher for the early than for the later stages. *a*, *b* and *c* represent the pollen-sterile form and *d*, *e* and *f* corresponding stages of the pollen-fertile form.



## LINKAGE BETWEEN MORPHOLOGICAL CHARACTERS AND FACTORS FOR SELF-STERILITY

F. G. BRIEGER                      and            A. J. MANGELSDORF  
*Harvard University*                      *Bussey Institution*

The investigations of East and his co-workers, especially of East and Mangelsdorf (1925) on *Nicotiana Sanderae* hort., of Lehmann (1926), and of Filzer (1926) on *Veronica syriaca* and the interpretation by East (1926) of experiments carried on by Baur on *Antirrhinum Segovii* (1919) have led to a very simple interpretation of the hereditary behavior of self-sterility in higher plants. In all of these three cases self-sterility is due to definite Mendelian factors forming a series of multiple allelomorphs which affect the rate of growth of the pollen-tubes growing through the style into the ovary. Pollen tubes carrying a factor also present in the tissue of the plant, when used as female in a certain cross, are inhibited in their growth and do not normally reach the ovules to accomplish fertilization.

This explanation permits the connecting of self-sterility with other phenomena in which genetical factors cause selective fertilization (Brieger, 1926). Sterility factors in fungi show exactly the same behavior, permitting the union of unlike gametes only (Brunswik, 1924; Burgeff, 1924; Hanna, 1925; Kniep, 1922). Factors affecting pollen tube growth as found by Correns in *Lychnis* (1921) and in *Rumex* (1922), by Heribert-Nilsson (1921) and by Renner (1921) in *Oenothera*, by Jones in *Zea* (1924) differ genetically from the self-sterility factors only in causing deficiencies instead of complete inhibition of certain matings.

A special feature connected with the self-sterility factors in higher plants is that there is a difference in the results of reciprocal crosses when the two parental plants have one sterility factor in common, as first pointed out by East and Mangelsdorf (1925). If, however, the plants used in crossing have no sterility factor in common, as  $S_1S_2 \times S_3S_4$ , the results of reciprocal crosses are identical since there is no selective fertilization.

Naturally, any factor linked with the sterility allelomorphs must show a similar behavior. Such a factor is the basic color factor in *N. Sanderae* which we have called "C." Plants carrying "C" in either the homozygous or the heterozygous condition have a purple color at the base of the stem, colored flowers whose shade depends upon the other factors present, and brown, rounded seeds. Plants carrying the homozygous recessive factor (*cc*) have a green color at the base of the stem, white flowers, and white, angular seeds. Under certain conditions, these flowers may show a slight pinkish tinge. This factor was found to be linked with the sterility allelomorphs in such a way that about 18 per cent crossing over was observed in both types of gametogenesis (Brieger and Mangelsdorf, 1926).

If we cross two plants heterozygeous for the *C* factor, we expect normally a 3:1 segregation; but this result will be obtained only if no elimination of gametes takes place, as in a cross of the type  $S_1CS_{2c} \times S_3CS_{4c}$  and its reciprocal. In crosses where the two parental plants have one sterility allelomorph in common very different results must be obtained.

Let us first assume that two plants are crossed in which the dominant factor "*C*" is linked with the sterility allelomorph not common to both parents, for instance the cross  $S_1CS_{3c} \times S_2CS_{3c}$  and its reciprocal. The pollen tubes which function,  $S_2$  in the one cross and  $S_1$  in the other, carry the dominant allelomorph "*C*" except for about 18 per cent crossovers carrying "*c*." Half of the offspring produced by the latter, or approximately 9 per cent of the total offspring, will be white flowering instead of the 25 per cent of homozygous recessives normally expected. Actually 68 colored and 14 white plants were found, the deviation from expectation being about three times the probable error.

If, on the other hand, the sterility factor not common to both parents is linked with the recessive allelomorph "*c*" as in a cross of the type  $S_1cS_3 \times S_2cS_3C$ , the functioning non-crossover pollen tubes carry the recessive allelomorph "*c*." The offspring will consist half of colored and half of white flowered plants, since there is no elimination of eggs in the heterozygous *Cc* plant. In addition 18 per cent crossover pollen-tubes will function carrying the dominant "*C*" allelomorph and producing, therefore, only colored offspring. In the total offspring about 59 per cent of the plants will be colored and 41 per cent white flowering. The actual numbers obtained were 342 colored and 246 white flowering plants, which is in sufficient agreement with the expectation.

If, finally, in the two parental plants different allelomorphs of the basic color factor ("*C*" and "*c*"), are linked with the sterility allelomorph which is not common to both parents, as in the cross  $S_1CS_3 \times S_2cS_3C$ , reciprocal crosses must give different results. In the type of cross given above the offspring will consist of approximately 59 per cent colored and 41 per cent white flowered plants. In the reciprocal cross all but 9 per cent of the plants will be colored. No data from a cross of this type are available at present.

Corresponding deviations from a normal Mendelian segregation may appear if heterozygous plants (*Cc*) are backcrossed to homozygous recessive (*cc*) plants. If the latter are used as male parents in a given cross the normal 1:1 segregation must result, the elimination of gametes having no visible effect. Actually 743 plants were colored and 788 were white flowering. If on the other hand the heterozygous plants are used as males the elimination of some of the gametes must result in disturbed ratios. We have to distinguish the following two cases.

If the sterility factor not common to the two parents is linked with the dominant basic color factor "*C*," as in a cross  $S_1cS_{3c} \times S_2CS_{3c}$ , the offspring should consist of colored plants only except for about 18 per cent white flowering plants produced by the crossover pollen-tubes. The numbers obtained



were 1,062 colored plants (expected 1,025.8) and 239 white flowering plants (expected 225.2).

If, on the other hand, the recessive factor "*c*" is linked with the sterility allelomorph not common to both parents, as in a cross  $S_1cS_3c \times S_2cS_3C$ , all plants but the 18 per cent produced by the crossover pollen tubes should be white flowering. Of those obtained, 201 had white flowers (expected 214.4) and 69 colored flowers (expected 55.6).

It may be mentioned in passing that genetic analysis of *N. Sanderae* shows that the abnormal segregation found for the basis color factor "*C*" is not a general phenomenon characteristic of this material. Four other factors for flower color have been investigated, each of which shows a normal Mendelian segregation.

#### LITERATURE CITED

- Baur, E., 1919. Ueber Selbststerilität und über Kreuzungsversuche einer selbstfertilen und einer selbststerilen Art in der Gattung *Antirrhinum*. Zeitschr. f. ind. Abstamm. u. Vererbungsl. 21: 48-52.
- Brieger, F. G., 1926. Mendelian factors producing selective fertilization. Amer. Nat. 50: 183-191.
- Brieger, F. G., and Mangelsdorf, A. J., 1926. Linkage between a flower color factor and self-sterility factors. Proc. Nat. Acad. Sci. 12: 248-255.
- Brunswik, H., 1924. Untersuchungen über die Geschlechts und Kernverhältnisse in der Hymenomycetengattung *Coprinus*. Bot. Abhandl. 5: 1-152.
- Burgeff, H., 1924. Untersuchungen über Sexualität und Parasitismus bei Mucorineen. Bot. Abhandl. 4: 1-135.
- Correns, C., 1921. Zweite Fortsetzung der Versuche zur Experimentellen Verschiebung des Geschlechtsverhältnisses. Sitzungsber. d. preuss. Akad. Wiss. 18: 330-354.
- 1922. Geschlechtsbestimmung und Zahlenverhältniss der Geschlechter beim Sauerampfer (*Rumex acetosa*) Biol. Zentralbl. 42: 465-480.
- East, E. M., and Mangelsdorf, A. J., 1925. A new interpretation of the hereditary behavior of self-sterile plants. Proc. Nat. Acad. Sci. 11: 166-171.
- Filzer, P., 1926. Die Selbststerilität von *Veronica syriaca*. Zeitschr. f. ind. Abstamm. u. Vererbungsl. 41: 137-197.
- Hanna, W. F., 1925. The problem of sex in *Coprinus lagopus*. Ann. Bot. 39: 431-458.
- Heribert-Nilsson, H., 1923. Zertationsversuche mit Durchschneidung des Griffels bei *Oenothera Lamarckiana*. Hereditas 4: 177-190.
- Jones, D. F., 1924. Selective fertilization among the gametes from the same individuals. Proc. Nat. Acad. Sci. 10: 218-221.
- Knip, H., 1922. Ueber Geschlechtsbestimmung und Reduktionsteilung (Untersuchungen an Bastidiomyceten). Verh. d. Phys. Med. Ges. zu Würzburg, N. F. 47: 1-29.
- Lehmann, E., 1926. (These proceedings, in press.)
- Renner, O., 1921. Das Rotnervnenmerkmal der Oenotheren. Ber. Deutsch. Bot. Ges. 39: 264-270.



## WHY *LYSIMACHIA NUMMULARIA* L., ALWAYS VERY FLORIFEROUS, IS ORDINARILY STERILE

F. GAGNEPAIN

*Museum d'Histoire Naturelle, Paris*

I have never observed in nature the fruits and seeds of *Lysimachia Nummularia*, although this plant is very common in France and flowers profusely, nor have the majority of the French botanists seen the fruits of this plant. But the authors of "Flore des Deux-Sèvres" (III p. 157, Sauzé and Maillard, describe fruit and seeds: "Capsule globuleuse, à 5 valves; graines noirâtres, anguleuses, grossièrement chagrinées." Furthermore, several authors cannot tell in what manner this plant, so very common in wet places, propagates itself.

This explanation of the sterility of this species occurred to me: Perhaps this *Lysimachia*, like other plants that live in plethoric conditions, has no good pollen,<sup>1</sup> and my studies have confirmed this hypothesis. Six individuals were potted and put in the experimental garden of the Paris Museum and disposed at graduated levels, so that the pot of Number 1 was in a hole with its top at the ground level, and that of Number 6 was simply placed on the ground. Number 1 was in the wettest condition and Number 6 in the driest one and so on, the intermediate numbers in sequence in intermediate conditions of soil moisture.

These six individuals were equally watered during the experiment period from April 30th, 1922. They were not protected against the rain, which was frequent this year. The results of the experiment, on June 12th, were as follows:

- Number 1, Plethoric strong plant, six flowers only.
- " 2, Bad condition, poor plant; two flowers for experiment.
- " 3, Normally strong, thirty flowers.
- " 4, Normal, strong, twenty-four flowers.
- " 5, A little feeble, five flowers.
- " 6, Thirsty, three flowers.

Plant Number 3 seemed to be in the best condition. It is valuable to know whether (a) the stamens were good, with fertile pollen, and (b) the pistils were likewise fertile. Both were perfect. I expected to see under the microscope wrinkled pollen; I saw ovoid-oblong grains, pale yellow, the ends subtruncate with three longitudinal furrows; exine minutely areolate; and 99 per cent were turgid without any wrinkles. In brief, there was perfect virility. The pistil (ovary, ovules, style and stigma) was sound. Furthermore, style and stigma move in a very peculiar manner, going toward the

<sup>1</sup> Gagnepain, F., 1913. Le pollen des plantes cultivées. Bull. Soc. Bot. Fr. 60: 224-231.

stamens. This movement was described by H. Lecoq,<sup>2</sup> and I entirely agree with him.

The style is needle-formed and not at all rigid. One knows that, in this species, the leaves are opposite and spread on the ground. The axillary pedicels are oblique, being short and stout. They are oblique in two distinct ways, (1) 45° with the direction of the stem, being axillary, and (2) 45° with the vertical, being ascending.

Naturally the flowers have their circle oblique in corresponding manner. But the needle-formed style is very supple, it tends to the verticle and toward stamens of the side nearest the stem and in time touches one of them. If the flower is on the right side of the stem, its style touches the left stamen of the circle; on the contrary, if the flower is on the left side, its style touches the right stamen. So we understand that when the pollen grains are perfect the pistil is likely to be perfect, and the style has a movement that realizes the pollination. But pollination very rarely results in seeds.

In my experiments, I expected to obtain fruits and to learn why under natural conditions fruits are not produced. In 1922, June 19th, pollination was tried on several opened flowers by means of a pencil. The day after, I cut out three branches so that the sap was concentrated in the opened flowers which were respectively 1, 5 and 6 in number. On June 27th, many pollenized flowers had lost their corollas, their sepals closely enveloped the ovary and their pedicels had bent down. Evidently maturation had occurred.

On July 13th the results of the experiments were as follows:

Numbers	Flowers	Fruits on the whole	Cut Branches	Percentage of fruits
1	6			
3	30	0		0
4	14	13		46
5	4	5	4	35
6	3	3		75
		2		66

Numbers 1 and 2 yielded eight flowers but no fruit.

Numbers 3, 4, 5 and 6 which presented no plethoric conditions yielded respectively 30, 14, 4 and 3, or the total of 51 flowers yielded 13, 5, 3 and 2 fruits—a total of 23.

On plant Number 3, three cut branches gave 22 flowers and 13 fruits. On the same plant, uncut branches gave 5 flowers and 4 fruits. (The result of cutting branches is not good.) In wet soil individuals gave few flowers and no fruit. The extreme dryness of soil does not favor flowering but does favor fructification.

In nature, at Chaville, between Paris and Versailles, I found plants of *Lysimachia Nummularia* growing under natural conditions that are of interest. On July 20th, 1922, many individuals were in blossom with the ovaries beginning to enlarge. The year before it was the contrary in the same spot. At that time, the trees of the forest had recently been cut out and there were then

<sup>2</sup> Lecoq, H. *Géographie botanique* 8: 125-126.



no grasses on the wet soil. All the individuals had flowers in large quantities, but no fruit appeared in any of them. In 1922, the same locality had the appearance of a meadow before the coming of hay-makers. The individuals of *Lysimachia*, instead of creeping and rooting on the ground, had grown up on the grasses. The soil was dessicated by the many grass roots. Many ovaries were developing into fruits. Perhaps the inability of creeping and rooting had favored the production of fruits. Was the production of fruits favored by dryness of soil? Was it the inability of rooting and creeping which favored the production of fruits?

From that moment, I thought it was useful and necessary to proceed to new experiments. In 1925, I had in my own garden at Montgeron, between Paris and Fontainebleau, a stout tuft of *Lysimachia Nummularia*, about a square meter in area. The spot was shady and the soil was deep and rich. I decided to place under opposite conditions the two halves of the tuft, putting the left part on an iron net so that it was impossible to creep and root on the ground. The right side could, on the contrary, make branches and leaves freely on the soil and form adventitious roots. The whole was in season profusely flowered on the left and right parts, but in no case did an ovary begin to grow and no fruit ripened.

It appears after the results of the 1925 experiments that the dryness of soil in my experiments of 1922 produced fertility in the individuals numbered 3, 4, 5 and 6, and that it was not merely the ability to creep and root which had permitted the development of fruit. In my 1925 experiments, plethoric conditions were permitted by moisture of soil, shade, etc., like in nature.

Another species in the French climate very often has no seeds. It is the *Ficaria ranunculoides* L. which flowers very profusely. But in nature, it happens that achenes ripen when the individuals are in well-drained soil, or when at flower season one digs up the plant and puts the individual in a dryer spot.

The propagation of *Lysimachia Nummularia* is not well known by botanists. Ch. Royer himself, who studied with such accurate attention the radical system of plants, gives no sufficient explanation.<sup>3</sup>

My observations are as follows: *Lysimachia Nummularia* produces, in many directions, creeping branches that are very often floriferous. Near the end of branches, adventitious roots are born at nodes. Then one to three buds grow into as many small subverticillate branches. The flowers are shed and the principal branches decay. In winter one can see only one to three small terminal branches which are still living and which will propagate the individual. By this means the plant can advance in a year to a distance of 30 to 60 cm. in every direction.

This point determined, it seems interesting to know the seedlings of *Lysimachia Nummularia* which are yet unknown, the seeds being very rare. The seeds from our many fruits were sown on the 24th of February, 1922.

<sup>3</sup> Ch. Royer. Flore de la Côte-d'Or 1: 226.

Three months later the seedlings were well developed and had four nodes and were four centimeters high. A description of them is as follows: Stem under cotyledons, a little oblique, 6 mm. long. Cotyledons elliptic, 6 mm. long, 2.5 mm. wide. Leaves spreading, opposite decussate; first node obovate, subelliptical, a little attenuate at base in a short petiole (1.5 mm. long) 10 mm. x 5; second node 13 mm. long, 7 mm. wide, distinctly obovate; third node widely elliptic, 15 mm. long, 8 mm. wide with 3 to 4 pairs of secondary nerves; fourth node leaves young, obovate linear, erect, not full grown, 5 mm. long, 2 mm. wide.

It is interesting to note that the stem is erect, the leaves decussate node by node, and that the leaves are never orbicular as in full grown plants. Of course, the erect stem must in time spread on the ground, or the axillary buds must spring up, grow quickly, and begin to creep on the ground. It must be said that the latter explanation is more reasonable, for axillary buds begin to grow and develop at a very early stage.

The *Lysimachia Nummularia* case resembles those of *Lilium candidum*, *Allium sativum*, *Artemisia dracunculus*, *Ficaria ranunculoides*, etc., which have no fruits. This sterility is probably produced by a predominance of the vegetative system on the gametic system.<sup>4</sup> This predominance involves plethoric conditions which are not the same as many different species are concerned, because every species has its proper temper.

If one can reduce in peculiar manner this predominance, one can often obtain fruits on a very sterile plant. The difficulty is to apply the measure and the proceeding.

#### SUMMARY

*Lysimachia Nummularia* is profusely flowering, but very rarely produces fruits under natural conditions.

It may be asserted at first that both stamens and pistils are sound and perfect.

To determine why this plant is sterile and to obtain fruits and seedlings from it, six individuals were cultivated in as many graduated conditions of moisture. Twenty-three fruits were obtained.

Two explanations may be given for this result: (1) Was the production of fruits favored by dryness of soil? (2) Was it the inability of rooting and creeping (in experiments) which favored the production of fruits? Experiments were made which answered the first question in the affirmative.

The author concludes by giving the way by which *Lysimachia Nummularia* propagates in an agamic manner. A description of a seedling of this species and general views on the question of plant sterility are given.

<sup>4</sup>Compare F. Gagnepain, Le pollen des plantes cultivées. Bull. Soc. Bot. France 1913: 224.

## CONCERNING THE STERILITY OF PHANEROGAMIC PLANTS (FRENCH STUDIES)\*

D. BOIS

*Museum d'Histoire Naturelle, Paris*

Sterility in plants should be combatted since it destroys or reduces more or less the production of the fruits and seeds that are indispensable to man and necessary for the multiplication of plants by seed and for the improvement of races by means of hybridization. Sterile plants, however, may have utility; certain sterile trees are sought for planting along public or other particular walks. This is the case especially with the common double-flowered chestnut (*Aesculus Hippocastanum* var. *flore pleno*) and the red chestnut (*Aesculus rubicunda*) preferred to the common chestnut with simple flowers which is very disagreeable for pedestrians during the period of the shedding of fruits.

A suppression of flowers on young trees is the current practice in horticulture, to prevent premature fructification which would exhaust them and the practice is extended to the adult fruit trees in order to assure a balance between vegetative parts and production with as perfect as possible development of reserved fruit. In garden culture the suppression of inflorescences allows intensive development of leaves destined for food consumption, and the gardner hopes similarly to obtain an abundance of leafage of certain species which are thus made more ornamental. In certain cases this permits also of the prolongation of life in certain monocarpic plants. The petalody of the *androecium* and of the *gynaeceum* is also, a peculiarity which man has always sought to develop in flowers to make them more ornamental as shown in the considerable number of double or full-flowered varieties known today. Finally, it is known that aspermous fruits are at times appreciated and that growers try to preserve this characteristic in bananas, pineapples, bread fruit, oranges, etc.

### CAUSES OF STERILITY

They are numerous and may be summarized as follows: Insufficient or excess of warmth, of light or of moisture in the soil or in the atmosphere; meteorological accidents; insufficient or excessive food; separation of sexes as seen in dioecious plants; self-sterility, atrophy or peculiar constitution of organ of reproduction; cryptogamic disease, and injuries by various animals.

---

\* The translation of this article from French to English was made by Dr. Illo Hein of the Department of Botany, Columbia University.

## LIFE OF PLANTS ACCORDING TO THEIR GEOGRAPHIC DISTRIBUTION, NATURALIZATION AND ACCLIMATIZATION

Each region of the globe, taken by itself, would be poor in economic and ornamental plants if it were limited only to the indigenous flora. Man is also inclined to the introduction of useful plants, first from neighboring regions and later from more distant countries when the means of communication permits it. The word "acclimatization" in its limited meaning is applied to a thing very difficult if not impossible to realize since it would consist in adapting a being to a climate distinctly different from that of its native country in such fashion that it can not only live but propagate naturally in an indefinite manner without the help of man. The cases of acclimatization generally cited are cases of "naturalization." That is, they are concerned with plants that have been transported to countries more or less distant from those from which they originated and have found there conditions of climate sufficiently favorable to permit them to prosper and multiply like the indigenous species.

Certain plants have a limited geographical distribution with special requirements which render them difficult to transplant elsewhere. Others, on the contrary, have an extended area of distribution and at times present natural races adapted to relatively variable climates. Finally there are ubiquitous forms (*Poa annua*, etc.).

As examples of naturalization in France one might cite, *Robinia Pseud-acacia* L., which originated in North America. The first plant of this species introduced to Europe still lives in the Jardin des Plantes de Paris. Today this species is abundant in temperate parts of the old world. Among the other American plants which grow in a subsontaneous state may be mentioned, *Erigeron canadensis*, *Elodea canadensis*, *Agave americana*, *Opuntia Ficus-indica*, etc. These have spread to such an extent in the Mediterranean region that they seem to be characteristic of the flora. A large number of useful and ornamental plants have been propagated in this manner outside their own country.

When the change in climate is too abrupt, life is impossible to perennial or to woody plants. If the change is less marked existence would be accompanied with reduced vitality, with the absence of flowering or the more or less complete sterility of the flowers. It is thus, therefore, that we possess in botanical gardens and in private collections numerous exotic plants that are infertile. Annual plants escape this rule in a region with a summer warm enough and long enough to permit them to entirely complete their cycle of development. The plants of colder regions introduced into warmer countries may live provided the difference in climate is not too great. But the absence of clearly marked seasons may keep them in a state of continuous vegetation which is unfavorable to fructification to the point where it may be abolished. This happens, for the most part, to fruit trees from temperate climates which have been introduced into tropical countries.

In nature a well-developed plant living in surroundings to which it is adapted finds itself under conditions which assure its fertility to the utmost.



However, it can become sterile if the factors which operate to insure its complete development are unfavorably influenced.

As Mr. Lucien Daniel<sup>1</sup> says, that it is very rare in nature for a plant to be surrounded by an entirely favorable environment. It is nearly always placed in surroundings varying with countries and seasons. The relative composition of the soil, water content, light, etc., are often imperfect but may be modified artificially by man.

The absorbing and aerating apparatus of a plant is generally so disposed that when these variations of soil and climate are only moderately different from those in which the plant naturally lives there are only temporary passing injuries. In which case the plant, after a more or less prolonged struggle in establishing the equilibrium of nutrition, ends by creating for itself a new *modus vivendi*, a temporary adaptation to the surrounding environment.

In a dry medium, the plant reestablishes its nutritive equilibrium by reducing transpiration through the shrivelling or shedding of its leaves, or by a production of rootlets or a lengthening of the root which can in this manner attain lower depths in the soil where there is more moisture than at the surface. In case of greater drought, which is more dangerous, the plant reduces its organs to the minimum size necessary to carry on active life in order to pass rapidly to a more quiescent state in the form of seed or rhizome, its sexual maturity is advanced, the fruits are less numerous and smaller, its resistance to exterior agents is lowered and its life shortened.

In a medium highly favorable to life, the tap root destined for the lower more moist strata disappears and the root becomes fascicled with numerous small roots which develop in the upper layers of soil. The elaborated foods no longer go to the roots as in the preceding case, but toward the stem which acquires new green organs of transpiration. The production of fruits and seeds is increased but the seeds are smaller and tend rapidly to the degeneration of the race. There is then a weakening of the race under the influence of humidity by an increased growth which at first appearance seems to favor the individual. Similarly, resistance to exterior agents diminishes especially in regard to cryptogamic diseases. This greater susceptibility is accompanied furthermore by a shortening of the length of life.

The abortion of certain organs in plants is frequent and botanists often mention it in their works. It is thus that ovaries normally having several carpels at the time of flowering have but one when the fruit is mature, others having been obliterated. It also happens that in the carpel or carpels that do persist all the ovules that existed in the ovaries did not become seeds. These abortions may be noticed for example in the cocoanut (*Cocos nucifera* L.) in which the ovary possesses three carpels, two of which become obliterated; in the date palm (*Phoenix dactylifera* L.) there are three carpels of which only one becomes the fruit; in oaks the ovary has three carpels each containing two ovules but the fruit (acorn) has but a single seed. These peculiari-

<sup>1</sup>La théorie des capacités fonctionelles et ses conséquences en agriculture. Bull. Soc. Sci. et Med. de l'Ouest, IV trimestre, 1902.

ties may be observed in other plants such as in nut trees (*Corylus*), the chestnuts (*Castanea*), etc.

Very frequently seeds which appear normally developed lack embryos for various reasons. The proportion of these seeds without embryos is often very large among seeds of the cultivated trees and shrubs grown in France.

In nature, reproduction in a plant instead of being sexual may become asexual. (*Agropyrum repens*, *Ammophila arenaria*, *Elymus arenarius*, *Lysimachia Nummularia*, *Phalaris arundinacea*, *Ranunculus Ficaria*, *Vinca major* and *V. minor*, etc.) The exaggerated development of a root system or of radican branches totally or partially prevents the production of seeds.

It is known that the ivy (*Hedera Helix*) which is bloomless while the branches creep on the ground blooms and fructifies on the specialized branches which develop when the plant creeps on a support. The characteristics of plants which fruit on specialized branches are not peculiar to this plant. They may be found clearly characterized in *Ficus repens*.

M. Gagnepain, Assistant in the Museum, has observed that the celandine (*Ranunculus Ficaria* L.) loses its power to produce numerous bulbils and fruits when it lives in a relatively dry medium. He obtained the fructification of *Lysimachia Nummularia* L. in the experimental gardens of the Museum in Paris by transforming by certain artifices the radican stems of this plant into upright stems which could no longer fasten themselves in the soil.

In the cultivated state certain plants produce only atrophied flowers, as has been observed in France for *Stachys affinis*. I have been able, however, to testify to a normal flowering of this plant.<sup>2</sup> The floral reduction is occasionally very great in certain potato varieties and this has been similarly observed in the sweet potato (*Ipomoea Batatas*) which is sometimes totally lacking in seeds.<sup>3</sup>

This peculiarity is still more accentuated in common garlic (*Allium sativum*) which hardly ever fruits in France, in *Artemisia Dracunculus* L. in *Oxalis Deppii* Lodd and in *Oxalis cernua* Thunb.

In other cases the number of flowers is so great in the inflorescences and the seeds so small and so rare that certain plants have been looked upon as sterile, as for example, sugar cane (*Saccharum officinarum* L.). It seems that Rouf of Martinique observed in about 1879 the first sugar cane grown from seed. This observation was published<sup>4</sup> and it was cited also by Boname<sup>5</sup> in his work on the sugar cane of Guadelope. According to Henry de Vilmorin<sup>6</sup> the first sowings of this plant were made at the Samarang station (Java) by Soltwedel.

Under Parisian climate, sterility of certain plants is apparently due to

<sup>2</sup> Revue Hort. 63: 463. 1891.

<sup>3</sup> Sagot. Jour. Soc. Nat. Hort. France 1871: 150.

<sup>4</sup> Deuxième mémoire sur la Canne à Sucre. 1888.

<sup>5</sup> Landes. L'Avenir de la Canne a Sucre a la Martinique. Rev. Cult. Colon. 1901: 257.

<sup>6</sup> Les semis de canne a sucre. Rev. Cult. Colon. 1897: 166.

the fact that their flowering period occurs at the time when there is great decrease in temperature. Too precocious are *Forsythia* (different species), *Jasminum nudiflorum* Lindl., *Lonicera fragrantissima* Lindl. and Paxt., and *L. Standishii* Hook., *Camellia* (the different species), *Parrotia persica* C. A. Mey., *Prunus Davidiana* Franch., *Stachyurus praecox* Sieb. & Zucc.: too late are *Zizyphus sativa* Gaertn., *Boussingaultia baselloides*, *Caryopteris Mastacanthus* Schauer, *Plumbago Larpentae* Lindl., *Ilex* (different species), *Abelia chinensis* R. Br., *Dielytra spectabilis* G. Don., *Oxalis crenata* Jacq., *Plagiospermum sinense* Oliver, *Ribes multiflorum* Waldst & Kit. *Carya olivaeformis* Nutt. fruits only in the south and west of France and grows without producing fruit in the center of this country.

In Paris itself at the Museum (Jardin des Plantes) where the conditions for existence are especially poor for plants, cases of sterility are numerous. Among the most characteristic cases may be cited *Acer creticum* L., *Apios tuberosa* Moench, *Berberis stenophylla* Masters, *Cedrela sinensis* Juss. (which fruits rarely), *Cedrus Libani* Barrel, *Corylus Colurna* L., *Fraxinus excelsior* var *monophylla*, *Kniphofia aloides* Moench, *Lilium candidum* L., *L. croceum* Chaix., *Liquidambar orientalis* Miller, *Phillyrea decora* Boiss. and Balansa, *Polygonum Baldschuanicum* Regel, *P. cuspidatum* Sieb. and Zucc., *Prunus tomentosa* Thunb., *Quercus macrocarpa* Michx. (of which there is a large infertile specimen), *Rubus deliciosus* Torr., *Rubus odoratus* L., *Uvularia grandiflora* Sm., *Zelkova crenata* Spach. (large trees which never fruit), etc.

I shall omit without comment those plants whose sterility is due to the peculiar conformation of their flowers and whose fecundation is in nature due to the intervention of insects as in the majority of exotic and indigenous orchids, and also in *Aspidistra elatior* Blume, etc.

A sudden increase of water near a plant would explain the appearance of teratological cases in the same way as has been observed by M. Camus.<sup>7</sup> The changing of male flowers to female flowers is associated in other plants with an excess of food or with injury as in *Zea Mays*, *Carica Papaya*, *Cannabis sativa* L., etc.

Monoecious examples of dioecious palm trees have been described by M. Friedel<sup>8</sup> for *Trachycarpus excelsa* Wendl, and by Bois<sup>9</sup> for the date palm.

But plethora, which is so frequent in cultivated plants, may bring about other teratological modifications injurious to fertility, such as fasciation of stems and inflorescences, phyllody, chloranthly and petalody or doubling of flowers. Intensive culture is particularly unfavorable as is attested by numerous examples.

Remarks concerning this subject made on Chrysanthemums cultivated to very large flowers show that the monstrous heads of these plants give flowers in which the organs of reproduction generally are atrophied.<sup>10</sup>

<sup>7</sup> Bull. Soc. Bot. France 51: 140. 1904.

<sup>8</sup> Bull. Soc. Bot. France 54: 192. 1907.

<sup>9</sup> Rev. Hort. 82: 492-494. 1910.

<sup>10</sup> Chiffot. De la production des graines de Chrysanthème. Rev. Hort. 1908, p. 39.



M. Chiffot cites among the varieties cultivated to large heads which cannot be fertilized, Reine d'Angleterre, Mistress Harman Payne, President Lemaire, Beauté Lyonnaise, André Charmet, Le Chalonnais, Soleil d'Octobre, Gustave Henry, Philomène Claret and Madame Chevraut, while the same varieties cultivated for small heads have well constituted female organs. It is understood that in the monstrous heads the stamens have all been transformed to sterile foliaceous organs, making it necessary to seek pollen from less deformed heads which have preserved their anthers. The varieties Rayonnant, Waldeck Rousseau, Madame Edouard Rey and Coletta also cultivated to a large flower always have normal pistils.

In all plants the duplicated part is generally the androeceum, rarely the gynaeceum. Brongniart<sup>11</sup> has, however, observed an example of pistilody of ovules in *Primula sinensis*. Another case has been described by Baillon<sup>12</sup> in *Sinapis*; then another by Vuillemin<sup>13</sup> on *Begonia erecta*. Transformation of floral organs to carpels is seen in *Cheiranthus Cheiri* L. Examples have been cited among diverse *Begonia*, notably by D. Bois.<sup>14</sup> Another particularly remarkable case has been observed by Duchartre.<sup>15</sup> But stamens may undergo less extensive modifications not always revealed to the naked eye but of capital importance from the point of view of their role.

The relatively short (one to five days) duration of germination of normal pollen of certain plants as *Cerastium vulgatum* L., *Lamium album* L. and *Rumex Acetosella* L. and the much longer period for others such as 55 days for *Vinca minor*, 70 days for *Agraphis nutans* and 80 days for *Narcissus Pseudo-Narcissus*, etc., have been established by L. Mangin.<sup>16</sup> They may likewise have a still longer period of germination (see the special discussion of sterility in orchids).

Under the influence of diverse poor conditions in nature and above all under cultivation, especially under hybridization, pollen grains may be completely altered in constitution and completely atrophied while their anthers have preserved a normal appearance.

We owe to M. Gagnepain<sup>17, 18</sup> two notes on sterility of pollen, one on that of hybrids, the other on cultivated plants. They show that pollen of wild plants is not always totally fertile as is generally believed, that cultivated plants present a fertility noticeably reduced because of either more or less defective conformation, and that partial sterility of pollen as a criterion of hybridity should be rejected.

In his thesis "Recherches botaniques sur les variétés cultivées du *Sola-*

<sup>11</sup> Ann. Sci. Nat. Bot. II. 1: 308, 1834, and III. 2: 20, 1844.

<sup>12</sup> Adansonia 3.

<sup>13</sup> Bull. Soc. Bot. France 42: 143. 1895.

<sup>14</sup> Jour. Soc. Nat. Hort. France III. 21: 949-956. 1899.

<sup>15</sup> Jour. Soc. Nat. Hort. France 1879: 171.

<sup>16</sup> Bull. Soc. Bot. France 33: 337. 1886.

<sup>17</sup> Bull. Soc. Hist. Nat. Autun 1901: 20.

<sup>18</sup> Bull. Soc. Bot. France 62: 224. 1913.



*num iuberousum*," 1911, P. Berthault presented the results of his examination of pollen of numerous varieties of potatoes of which the flowers were often unfecundated because of the poor constitution of pollen. But the action of normally constituted pollen is hindered or destroyed by meteorological accidents, such as abundant rain, lowering of temperature, excessive atmospheric dryness, etc., which prevents its arrival to the stigma or destroys it before fecundation has taken place.

Self-fertilization is the rule in cleistogames. But in many flowering plants the floral organs may be of such a conformation that pollination is rendered impossible without special intervention. This is the case with the greater part of the orchids, of diverse Labiatae and Scrophulariaceae, etc., in which sterility is absolute or at least partial unless the pollen is carried to their stigmas by insects or by artificial pollination. The case of vanilla (*Vanilla planifolia*) in which fertilization is assured in its native country but which remains sterile in Reunion where it was introduced for cultivation, is a striking example of the pollination by insects. It is known that artificial pollination by the hand of man results in fructification and it is now practiced in a routine manner not only for vanilla but also for other exotic orchids cultivated in our greenhouses.

Numerous experiments made upon animals and plants have established that the union of cells belonging to closely related plants is prejudicial to the conservation of organisms. It is frequently a verified fact that certain plants cultivated separately are on this account more or less sterile (self-sterile plants) in that case they are fertile when other plants of the same or similar species or variety which have the same period of flowering and good pollen are grown in their vicinity. However, that may be, cross-pollination is the rule in dioecious, monoecious and self-sterile plants because of the peculiar formation of normal and abnormal flowers. It is thus very common in nature as noted by Lecoq.<sup>19</sup>

### STERILITY OF EXOTIC PLANTS CULTIVATED IN FRANCE

I owe to M. Georges Poirault, director of Villa Thuret, Laboratoire des Hautes-Etudes, Ministère de l'Instruction publique, cap d'Antibes, the observations given in this section, more or less directly quoted from his statements.

Many exotic plants grow very vigorously and flower, but do not fruit or fruit only occasionally in the Villa Thuret. I shall leave aside the plants to which pollination is effected by insects which are lacking in our fauna as the *Promubas* which pollinates certain species of *Yucca*.

Sterility may be due to a malformation of pollen or ovules (malformation due perhaps to temperature at the moment of formation of the sexual elements). All our sages (*Salvia*) from Mexico are sterile. Perhaps I could make these plants flower at a time when the question of temperature would be eliminated. In any case this is not a fact. Sterility on account of malforma-

<sup>19</sup> Comp. Rend. Acad. Sci. 1862: 1247; and Bull. Soc. Bot. France 9: 211. 1862.

tion may again be general (all the flowers of *Theodora* are anomalous). All *Araliacées* are apparently sterile because of malformation of the stigma. There are, on the other hand, plants whose flowers set fruit only at a certain time, flowering may be extremely abundant, but flowers generally drop. If proterandry is too marked it is possible that the pollen will not be in a state of germination when the stigmas are receptive.

Certain plants (*Elacodendron croceum*, *Arganis Sideroxylon*, etc.) flower only occasionally but then give many fruits with fertile seeds. *Berberis nepalensis* flowers annually but I have seen it set fruit only once in twenty-six years. *Lithraea venenosa* blooms often but rarely sets in abundance. We formerly distributed seeds of *Lagunaria Patersonii*, which indicates that the seed should be abundant enough, but for four years at least all the seeds of one plant were abortive.

It will be necessary to note again the persistent influence of cold on plants which were subjected to freezing. For years *Corynocarpus laevigatus* bloomed regularly without ever fruiting, but since the severe frost of 1920 not a single flower was produced. *Ficus Hookeri*, severely hit by the frost of the same year, remained four years without flowering. It resumed feebly last year. Like the previous ones the seeds appeared sterile or at least I have not known them to germinate. *Jacaranda mimosifolia* flowers very abundantly but there is perhaps not a single flower in a thousand which sets. The chief factor in all this matter could very well be temperature.

### PARTHENOCARPY

In normal cases the fertilized ovules develop to form seeds at the same time that the walls of the ovary develop and together they form the fruit. But it may happen, for one cause or another, that although the ovules are not fertilized the pericarp develops fruit having all the appearance of a good constitution but is aspermous. Examples of this are seen in bananas, pine-apples, seedless pears, seedless oranges, Corinth grapes, aspermous bread fruit, pomegranates, sapota, etc.

Naudin<sup>20</sup> sought to give an explanation of this phenomena and realized it by experimenting with various Cucurbitaceae, "Observations sur l'accroissement de certains ovaires et de leur conversion en fruits sans développement de graines embryonnées."

In 1902 Noll used the term "parthenocarpie" to designate this abnormal asexual reproduction which is occasionally established in varieties. Winkler<sup>21</sup> sees in this domain two types of behavior:

1. Parthenocarpy stimulative, determined by contact with the stigma of any pollen, or by an insect sting or most any other cause of stimulation.
2. Vegetative parthenocarpie, in which the sterile fruit is formed without pollination or of any excitation.

<sup>20</sup> Comp. Rend. Acad. Sci. 1857: 383.

<sup>21</sup> Ueber Parthenogenesis und Apogamie im Pflanzenreich. Prog. Rei. Bot. 2: 293-454. 1908.

## PARTHENOGENESIS

In 1694 Camerarius expressed the idea that seeds could be produced in spite of the suppression of male flowers and this question has become the subject of numerous studies. Van Tieghem considers that this phenomenon shows that one cannot regard sexuality as necessarily entailing sterility of isolated gametes, nor should we include, as is ordinarily done, this condition of sterility in the same definition with sexuality. Parthenogenesis, says this author, has, in short, the result of replacing sexuality prevented or lost by multiplication.

*Coelebogynne ilicifolia*, which is unable to reproduce itself on account of lack of pollen, substitutes an adventitious embryo for a normal embryo. *Pteris cretica* develops on its prothallium an adventitious embryo in place of the absent archegonium and thus the same general result seen in parthenogenesis is attained, but in another manner. It is obtained in a still different fashion in the umbel of *Allium sativum*, etc., in which bulbils are formed instead of flowers.

In all these examples the same results being obtained by different routes, sexuality is prevented or lost and is replaced at more or less distant points from those where they ordinarily operate, by different modes of multiplication. It is useful to have a common expression to designate this substitution. The term is apogamy. M. Pierre Lavielle<sup>22</sup> in his thesis on aggregation has given an extended list of publications which treats this question. •

The production of bulbils instead of flowers is found again in plants called "viviparous" as in *Fourcroya*, *Poa bulbosa* var. *vivipara*, different species of *Allium*, etc.

## STERILITY OF HYBRIDS

The study of hybridization and hybrids has permitted the obtaining of a considerable number of new plant forms, of which many are cultivated for beauty and utility. Duchartre<sup>23</sup> recalls that shortly before 1719 an English gardener, Thomas Fairchild, performed the first cross-pollination. After him, Linneaus, and above all, Koelreuter, made various experiments which gave a complete demonstration of the reality of this phenomena and made known the conditions as well as the consequences.

Charles Naudin is one of the botanists who has devoted himself to the most intensive and most persevering researches on this question. His researches covered several years of work in the Jardin des Plantes de Paris (Museum National d'Histoire Naturelle) with diverse plants. The results were published in a memoir<sup>24</sup> which earned the Prix Bordin of the Academie of Sciences for 1862. This was followed by later publications.<sup>25</sup>

<sup>22</sup> La Parthénogénèse chez les végétaux. 1914.

<sup>23</sup> Rapport sur les progrès de la botanique physiologique Paris. 1868.

<sup>24</sup> Observations concernant quelques plantes hybrides qui ont été cultivées au Muséum. Ann. Sci. Nat. IV. 9: 257. 1858.

<sup>25</sup> Nouvelles Archives du Muséum. 7: 25, 1865, and Ann. Sci. Nat. IV. 19: 180, 1863.



It is important to know whether the hybrids of two distinct species can reproduce themselves in their turn by fertilization or whether they are condemned to an absolute sterility. For a time after Koelreuter, physiologists maintained the latter of these ideas. The facts were differently presented by Naudin, for of 38 to 40 hybrids of species which he obtained and described, nine or more were sterile and three-quarters of the total number gave embryonic seeds which germinated with difficulty. Naudin noticed at the time the marked tendency of fertile hybrids to return toward the types which produced them at the end of a small number of generations.

Godron<sup>26</sup> had a totally different opinion from that of Naudin and which rested on two principles: first, species hybrids are constantly and absolutely sterile; second, hybrids sterile among themselves may be pollinated with one or the other parent, becoming fertile with them and thus give birth to quadrans endowed with more and more pronounced degree of the faculty of reproduction by seed.

In another memoir Godron<sup>27</sup> modifies his views and seems to approach the ideas of Naudin to which he was heretofore opposed.

Naudin has established that although some hybrids are absolutely sterile with stamens and ovary there are also some, and perhaps a very large number, which are fertile, some in respect to the ovary only, but others for both pollen and ovary.

M. Guignard was interested in determining the causes of sterility in plant hybrids in studying the development of the male organ and the female organ. The result of these learned and delicate researches were published in two memoirs<sup>28</sup>.

The male organ, he wrote, is perhaps stricken with different degrees of sterility, sometimes the stamens are transformed to staminoidia and there is no pollen. At other times the pollen grain stops in its development and dies before its nucleus is divided, although it may present a normal appearance even if its nucleus has not divided. In the last case (several *Begonias*) the grain germinates on the stigma forming a pollen tube but since it does not possess a generative nucleus it is unable to effect fertilization. The presence of two nuclei in a pollen grain does not always suffice to assure the faculty of fertilizing the female cell and its functional impotence might result in the loss of its normal characteristics before the dehiscence of the anthers.

The sterility of hybrids may also arise from the poor conformation of the female organ. Sometimes the number of ovules is less in hybrids than in pure species, but often on the contrary it is the same in the two cases. It is impossible to recognize simply by observation if the ovules which enclose the carpels of hybrid flowers are or are not capable of being fertilized. To be

<sup>26</sup> Ann. Sci. Nat. IV. 19: 135. 1863.

<sup>27</sup> Nouvelles expériences sur l'hybridité dans l'ère végétal. Comp. Rend. Acad. Sci. 1866: 379, and Mem. Acad. Stanislas 1865: 328-365.

<sup>28</sup> Sur les organes reproducteurs des hybrides végétaux: Comptes Rend. Acad. Sci. 1886 and Observations sur la stérilité comparée des organes reproducteurs des hybrides végétaux, Bull. Soc. Bot. Lyon, II. 4: 66. 1887.



settled on this point, says M. Guignard, it is necessary to see if the embryo sac is well developed. The absence of the embryo sac is one of the most frequent causes of the sterility of hybrids.

M. Guignard has similarly observed that the development of the female organ is not in constant agreement with that of the male organ. In the same flower the ovule may be very well developed and the pollen rudimentary or the reverse condition may exist.

In a manuscript which unfortunately remains unpublished, Edouard Bornet has given an account of the results of experiments on the hybridization in *Cistus* which he followed for ten years. In making more than 3,000 artificial pollinations, he established that plants of *Cistus* are nearly always physiologically dioecious, giving support to what we know today that, in many plants flowers can not be pollinated by their own pollen. In pure species of this kind fecundation fails because the pollen tubes cease to develop before arriving at the ovules or they do not penetrate to the ovarian cells.

In *Verbascum* hybrids the abortion of the capsule is remarkable because of its generality. A. Franchet<sup>29</sup> in a note having for its title, "Essai sur les espèces du genre *Verbascum* du centre de la France et plus particulièrement sur leurs hybrides," observed, however, that absolute sterility does not exist in these plants. Having examined from this point of view 600 hybrid individuals, he observed capsules only three times and one of these individuals was deformed.

In a note on the pollen of hybrids, M. Gagnepain<sup>30</sup> has shown that the pollen is often aborted in wild as well as cultivated plants. It cannot be concluded therefore that sterility is merely a mark of hybridity as some authors formerly affirmed.

### FUNGIOUS DISEASES AND STERILITY

Fungous diseases have at times a disastrous effect on plants for they may directly attack the whole reproductive organs as in the case of the cereal smuts (*Ustilago*), or the ovary of rye (*Claviceps*), or the anthers of certain Caryophyllaceae, an example of which has recently been observed in the gardens of the Museum of Natural History.<sup>31</sup> Certain virulent fungous diseases attack different parts of plants with a vigor that would reduce the fructification considerably.

### STERILITY CAUSED BY ANIMALS

Certain animals are destructive and it will suffice to indicate among them, "Phylloxera de la Vigne" (*Phylloxera vastatrix*) which lives on the roots of plants; Anthonomes, weevils whose larvae live in the buds of apple and pear blossoms devouring stamens and ovaries; butterflies whose larvae live in pears and apples causing them to fall prematurely from the trees, etc. There also are insects which on attacking the vegetative parts of plants cause monstrosities following defective nutrition.

<sup>29</sup> Mém. Soc. Acad. Maine-et-Loire. 22: 65.

<sup>30</sup> Bull. Soc. Hist. Nat. Autun 1901: 20-29.

<sup>31</sup> D. Bois, Revue Path. Veg. Paris 1923: 223.

## ANIMALS INSURING POLLINATION

Certain insects are essential to natural pollination of various plants.

There is a case of vanilla which was introduced in 1817 in the Island of Reunion where it remained sterile because the insect which insured its pollination in Mexico, its native country, does not exist there. Fruit production by this orchid is now regularly and easily obtained in practice by artificial pollination. The principle of this operation was discovered and applied in 1830 by Neumann, at Paris (in the Museum greenhouses) but his results were never published. Morren first spoke of it and its practical application started again in Reunion in 1841.

There is no longer occasion to insist on the role played by *Blastophaga psenes* which insures the transport of pollen from caprifios to the stigmas of female flowers of the fig tree (*Ficus carica*).

Authors in general attribute great importance to the part played by bees and other insects who pilfer the flowers and transport pollen from one to the other, thus insuring cross-pollination. There are those, however, who consider that its importance is exaggerated. In two notes Gustave Rivière and Georges Pichard<sup>82</sup> published the results of experiments in which ten pear and peach branches bearing unopened flower buds were introduced into paper sacks which were carefully closed to prevent the entrance of bees, butterflies, etc. On the same trees an equal number of flower buds which were just about to open were chosen and left in the free air under ordinary conditions to permit comparison. It is stated that in each case the number of fruit which set was the same and the conclusion made "que l'intervention des Abeilles ou autres insectes est absolument inutile pour opérer la fertilisation des fleurs de nos arbres fruitiers" (The intervention of bees or other insects is absolutely useless for the operation of fertilization in the flowers of our fruit trees).

## STERILITY IN GRAPES

In practice the name "coulure" designates especially the absence of development or incomplete development of seeds, but there are varieties of grapes, Corinth and Sultanieh (Sultanina) whose berries seem normally constituted, but which contain no seeds, the flesh having developed without fertilization. The sterility may be due to abnormal formation of flowers. Millardet designates under the name "hooded flowers" those in which parts of the corolla are united in a hood and because of a fixed character or of particular atmospheric conditions do not become detached whereas in normal flowers petal fall is induced by a straightening of the stamens which elevate the hood. The flowers called "long-stamened" have a perfect constitution and are always fertile, whereas the hooded flowers are often sterile. Millardet's studies have shown that the pollination of grapes is almost always crossed, although selfing may take place. There are also flowers with short stamens whose pollen is abnormal and incapable of germinating and whose normally constituted ovaries may develop into fruits after having been fertilized by the pollen of long-

<sup>82</sup> Jour. Soc. Nat. Hort. France 1923: 304, and 1925: 97.

stamened flowers or by pollen of male vines which have flowered with an aborted or a rudimentary pistil. The female flowers always possess stamens with filaments and anthers but both are in a rudimentary condition. They are fertile when pollinated by pollen brought to them by wind or insects.

Millerand or partial sterility of grape berries may be caused by a state of feebleness of vines, diverse maladies and by inclement weather. Lowering of temperature at the time of flowering, cold winds and cold rains are causes of accidental abortion which is also caused by dry winds which dry out the stigmas.

The grapes cultivated in greenhouses would be only slightly productive without artificial pollination when the outdoor temperature does not permit the opening of the ventilators for the purpose of pollination by wind or insects, and this proves that for grapes as for all plants in general, cross-pollination is necessary for the maintenance of the species.<sup>33</sup>

### STERILITY IN CIDER APPLES

Studies have been pursued by Mésenge de Beaurepaire and Ragaine for the finding of fertile varieties of cider apples on account of their more or less strong resistance to rough weather which occurs during blossom time. The results obtained by each of them have been published.<sup>34</sup> This number contains only a part of the observations of Mésenge de Beaurepaire, whereas the treatise by Ragaine<sup>35</sup> is complete.

G. Warcollier gives the essential part of these results in an abstract published in his book.<sup>36</sup>

The International congress of 1900 for the study of fruits for pressing has admitted, according to the calculation of Mr. Truelle, that a variety may be considered as fertile if its average production during at least ten years is equal at least to one-third of a full harvest.

Warcollier observed that such a definition is incomplete. In ten years apple trees may give one to three full harvests, then medium or small harvests and finally remain sterile for three or four years. It would be preferable, he says, for them never to give full harvests which weaken them but that they produce medium harvests regularly. The ideal, according to him, would be to have fertile varieties defined as varieties that yield harvests annually or at least have biannual harvests. He thinks that one could arrive at the result: First, by diminishing the number of flowers per year when these are too numerous, this may be accomplished by spraying with a solution of copper sulphate 0.5 or 0.3 per hundred; second, by trying to produce "in counter years," i.e., years in which all other apple trees have no fruits, by destroying all the flowers of the trees under experiment in the year when all apple trees are covered with them and where a rich harvest is foreseen. These trees

<sup>33</sup> Viala and Pacottet. *La fécondation artificielle de la Vigne*.

<sup>34</sup> *Bull. Assoc. Fran. Pom.* 17: gème fasc. 1899.

<sup>35</sup> The flowering period does influence the fertility of fruit trees.

<sup>36</sup> *Pomologie et cidrerie*. 2<sup>ème</sup> édition. Paris. 1920.



would thus be ready to give flowers and fruit the following year which would be most often a year when there is a scarcity of apples.

Certain producers, in order to arrive at the same result, observe and mark with conventional signs the apple trees which produce a full harvest during years of general weak production, and, if these trees yield regularly in counter years, grafts are made from them. But external circumstances such as hail, frost, June bugs, caterpillars, etc., may destroy the desired harvest and disarrange the established order causing the loss of the benefits from the operation.

Mr. Warcollier also believes that it would be possible to obtain regular annual harvests by cultivating, preferably, varieties whose flowers open only after the leaves have come out. With these varieties, flowers and young fruits are not exclusively nourished by reserves accumulated in the buds and shoots of the past season, but find the food necessary to the first development in the young leaves which elaborate it. Under these conditions, these varieties may be completely devoid of reserves after an abundant harvest, but are nevertheless capable of blooming and feeding the young fruits the following year.

The fertility of trees depends to a great extent on the conditions under which flowering took place, and this is at the mercy of good and bad influences, varying with the location of the orchards. The violent west winds break flowers and branches; those of the north and the east dry out the reproductive organs; late frosts of April and May also destroy many flowers; they are due to the intense nocturnal radiation which is frequent during the whole month of May or to the abrupt changes to low temperature, but the case is rarer. These frosts are especially to be feared in narrow deep valleys; in such locations, one should resort to late varieties. We are without means of defense against persistent rains which prevent the pollen from reaching the stigma, against downpours followed by intervals of strong sun which burns petals, stamens and pistils.

Because of meteorological accidents during the flowering period it has been sought to find whether there are periods during April and May when flowering is exposed to greater dangers than at other times in order that the cultivation of varieties which bloom at that time may be abandoned.

Mr. Mésenge de Beaurepaire grades the blooming of apple trees in five series:

- 1st—precocious, from the 15th to 30th of April;
- 2nd—semi-precocious, from the 1st to the 7th of May;
- 3rd—intermediate, from the 8th to 14th of May;
- 4th—semi-delayed, from the 15th to 21st of May;
- 5th—delayed, from the 22nd of May to June.

He has gathered numerous observations from 1,429 apple trees pertaining to 143 varieties. His conclusions are as follows:

Influences during the flowering period on the fertility of varieties are apparently the same and equally favorable for the first, fourth and fifth series.



No preference is to be established between them. The author advised restriction of cultivation to varieties pertaining to the second series.

Mr. Ragaine has devoted himself to the same studies for ten years and arrives at the following classification, in the order of their importance: fifth, fourth, second, first and third series. He concludes that the third flowering series (of the 8th to the 14th of May "Saints de glace") constitutes a critical period, and that it is desirable that trees which bloom before or after that period be cultivated.

Other growers condemn the cultivation of varieties of the first series, stating that they are often fruitless because of spring frosts and attacks by anthonomes, and recommend the planting of late flowering varieties.

It is also advantageous, Mr. Warcollier states, to propagate varieties whose flowers are rapidly pollinated and deflowered in a few days; because these flowers, being generally very fertile, have less to fear from adverse weather which is always dangerous if the flowers remain open for a fairly long time before pollination. Other varieties are interesting because flowering extends over a longer period, and the flowers come into bloom one after the other so that there are always some which are fruiting; these varieties are generally fertile, such as Améro, Gros Bois.

But the attention of the apple-growers should be directed especially to the necessity of selecting and propagating preferably those varieties whose flowers show the greatest resistance to adverse weather, and those in which pollination takes place even during days of north and west winds. Pomologists like Messrs. Ragaine, Hérisant, Cannet and Janneau have already made interesting observations regarding this subject; it has been seen that varieties such as Bonne-Cambrière, Cimetière, Fréquin du Mans. Bramtot which were fertile in 1907, flowered largely during rainy days in the spring of the same year.

VIGOR: The International Congress of 1900 admitted that a tree is strong if, after having reached full growth, it attains a diameter of 7 to 10 meters. During its development, says Mr. Warcollier, a vigorous tree may be recognized by its green foliage, rich in chlorophyll, by its tender, smooth bark, by the fact that it will give annually shoots at least 15 cm. in length and that it will grow continuously every year.

Strong, rapidly growing varieties should be cultivated but only on condition, however, that this rapid growth does not in any way impair fertility, and that its growth and fertility, on the contrary, advance evenly. Too vigorous varieties produce an abundance of wood, make their frame without giving fruit, remain sterile; these must be regulated or used as intermediaries. In a general way the very strong varieties should be put into soils which are least rich or least suitable for cultivation, and the less strong varieties into rich, deep soils.

According to Mr. Truelle, "resistance" of a fruit tree rests in its ability to oppose: 1st, atmospheric influences; 2nd, attacks by insects, and 3rd, invasion by canker and other parasites. One must, therefore, be careful that only varieties which are resistant to winter and spring frosts be introduced

into orchards and to take into consideration that in general varieties with soft wood are more sensitive than varieties with hard wood.

## STERILITY OF FRUIT TREES IN FRANCE AND IN MOROCCO

Knowing that M. Bey, Inspector General of Fruit Tree Cultivation in Rabat, Morocco, was interested in questions concerning sterility of fruit trees, I asked him to inform me of his observations. Concerning the records of these studies, he stated as follows: "Since I had a large nursery and valuable horticultural school at Marnay (Haute Saône) as a field of study, I had indeed become interested in research in sterility and fertility of fruit trees, and noted from 1906 to 1914 a great many observations and experiments, but the war came, interrupted my experiments and I was put to the painful task of abandoning my home and nurseries. The majority of my notes were lost at the time of our departure from Morocco, or were lost by my successor."

From his reports to me the following statements were obtained: A cherry tree variety, Belle Hortense, planted in company of some others produced magnificent and abundant fruits for some years. On account of a violent thunderstorm the cherry tree remained alone. Since then it flowered abundantly every year but bore only very little fruit up to the time when Kirsch cherry trees were planted and flowered in a nearby waste area. During this period of seven or eight years, the one remained quasi-sterile.

From this time on the flowers and pollen of a very great number of varieties of stone fruits and small fruits were studied and tested. Examination of the flowers immediately showed numerous cases of abortion (empty anthers, sterile pollen, flower abortion, etc.) which are much more frequent elsewhere than is generally imagined and the practice of artificial cross-pollination soon demonstrated the favorable influence of exchange of pollen from variety to variety. In the experiments, paper bags pierced by pin pricks were used, or sacs of fine gauze. Flowers were enclosed some time before their opening and later pollinated with their own pollen (self-pollination) or with foreign pollen (cross-pollination). When the fruits begin to set, and then again at maturity the percentage of fruits from the artificially pollinated flowers were determined and compared with the checks. Thus there was obtained after five or six years, a general idea of the sterility of certain varieties and the fertility of certain others, and also the favorable influence which certain varieties may have for the increase in production of certain others.

In spite of slight variations in the results from year to year, which are probably due to the age of the trees and for some of them the nature of the stocks, M. Bey was convinced that the great majority of our fruit varieties are not likely to produce abundantly by self-pollination, and that they are to a large part sterile, although they may be fertile in varying degrees among themselves and extended researches might establish that their degrees of sterility could be arranged in a graded scale. From the results the following tests were made:

## PEARS

*Self-fertile*—Fauvanelle et Belle de Berry, Conférence Triomphe de Jodoigne, Béguine (local variety), Nouveau Poiteau, Beurré d'Angleterre, Louise Bonne; then, at a less degree Bergamotte Espéren, Duchesse, Bon-Chretien Williams, Le Lectier, Beurré Diel, Beurré Clairgeau (on Coignassier), Passe-Crassane, Fondante des Bois (on Franc), Beurré d'Arenberg (on Franc), Duchesse d'Alençon, André Desportes, Beurré d'Amanlis.

*Self-sterile*—Pears considered as such since they give only three to six fruit settings per 100 flowers: Olivier de Serres, Beurré d'Anjou, Beurré d'Arenberg (on Coignassier), Beurré Clairgeau (on Franc), Belle Angevine, Passe Colmar, Charles Ernest, Beurré Superfin, Doyenné du Comice, Epargne, Assomption, Clapp's Favorite.

In Morocco, the varieties Epargne, Beurré d'Amanlis, Beurré Clairgeau, Souvenir du Congrès may be considered as sufficiently self-fertile.

Cross-pollinations made at random with a mixture of self-sterile varieties (by means of pen points which were not sterilized after each operation) resulted in increase of production of from five to eight per hundred. Others made with pure pollen (or at least considered as such) of known varieties, have still increased the percentage considerably. This was shown in the case of Olivier de Serres by Fauvanelle, Beurré Hardy by Beurré Superfin, Doyenné du Comice by Williams or by Conférence, le Lectier by Louise Bonne or Clapp's Favorite, and Beurré d'Anjou.

Since cross-pollination augments production, there would therefore be justification for the inter-planting of fruit trees according to the knowledge of the affinities of varieties. Perhaps it would prove interesting to seek in each region, a variety with a great strength of pollination which would be useful for planting in every orchard and every garden.

M. Bey states, "I shall not fail to recommend to all those of my clients who leave to me the care of planning their plantations to use as pollinating trees the following:

Pear trees: Fauvanelle, Conférence, Curé.

Apples: Double Rose, Baldwin, Queen of Russets, Calville du Roi.

Plums: Drap d'or de l'est Bégeonnières, Reine Claude violette.

Cherry trees: Planchoury, Guigne noire (Black heart cherry), Montmorency.

M. Bey classifies the principal varieties of apple, plum and cherry trees which he studied as follows:

## APPLES

*Self-fertile*: Api double (double rose), Châtaignier, Mottet (local variety), Baldwin, Calville du Roi (or King's Russet), Reine des Reinettes, Madeleine d'Été (Summer Madeleine), Reinette de Caux (Caux's Russet), Borovitsky.

*Self-sterile*: Sans Pareille (without parallel), Calville Lesans, Calville Saint-Sauveur, Ménagère, Ribston Pippin, Reinette Orange de Cox (Cox's orange Russet), Titovka.



*Slightly self-sterile*: Belle fleur jaune, Reinette d'Angleterre, Fleiner du Roi, Reinette du Canada, Reinette grise, Pomme Fraise, Jeanne Hardy, Reinette doree a Versailles, Grand Alexandre, Gravenstein.

### PLUMS

*Self-fertile*: Drap d' or de l'est, Damas, Bégeonnières, Reine Claude violette, (Reine) Claude de Bavay.

*Slightly self-fertile*: Mirabelle double, Perdrigon musqué (Musked Perdrigon), Quetsche d'Alsace, Reine Claude dorée.

*Self-sterile*: Monsieur Rouge, Belle de Louvain, Pruneau d'Italie, Prune Abricot, Pond's Seedling, Coe's Golden Relish, Reine des Mirabelles, Bleue de Belgique.

*Slightly nearer to self-sterility*: Prune d'Agen, Reine Claude précoce, Mirabelle petite.

### CHERRIES

*Self-fertile*: Planchoury, Guigne noire, Bechaà Kirsch, Montmorency, Bigarreau noir.

*Slightly self-fertile*: Carmélite, Impératrice Eugénie, Bigarreau Jaboulay, Bigarreau Napoléon, Gros Coeuret.

*Self-sterile*: Reine Hortense, Griotte, Guigne Garcine d'Olivet.

He states that cross-pollination greatly favors the production of stone fruit trees, plum as well as cherry trees. He states that cherries gave him strange contrasts from one year to the other and that his list should be taken only as a simple provisional indication. He regrets that it is the same for other lists.

At Morocco, says M. Bey: "I was particularly interested in the almond, for it is this tree that presents the most curious peculiarities in the various form of the styles in the flowers. It is a clear fact that the almond with long-styled flowers is pretty nearly the only very fertile one among the almonds in the region of Marrakech (perhaps also elsewhere).

Examination of the pollen of the flowers of almonds revealed to M. Bey that the greater part of the almonds in Morocco as well as in France had infertile pollen and the anthers were frequently empty. He found also that the round pollen grains (generally found in long-styled flowers) germinated easily while more or less elongated egg-shaped sinuate pollen grains were sterile.

From this it may be concluded—never plant by itself an almond tree which is not self-fertile; give preference to long-styled varieties; plant almonds close enough; plant a large number on a small place; raise bees for pollination, protect birds and floral beetles during the flowering season. It is to be noted that large fruited varieties such as Princesse sultana possess on the same tree many more long-styled than short-styled flowers.

### THE STERILITY OF ORCHIDS

Among the orchids in general self-pollination is impossible not only because the anthers and the stigmas are placed in such a fashion that direct pollination cannot take place, but also because the pollen instead of being pow-



dery and easy to disseminate as in most plant families is aggregated in masses (pollinia). In nature pollination is more or less assured by certain insects which transport the pollinia placing them in contact with the stigmas in successively pillaging the flowers.

Cross-pollination is the rule for exotic as well as for indigenous orchids. In orchids pollen has a double action, for it enables the growth of the ovary and the complete formation of ovules to take place before causing fertilization to take place. This behavior of pollen is clearly established by the remarkable studies of Guignard. In a preliminary note<sup>37</sup> he gives the results of observations showing that the ovules of these plants are still in most cases in a rudimentary state at the moment of pollination, and that the germination of pollen on the stigma and the penetration of the pollen tubes into the ovary are necessarily for their complete development.

A considerable time passes between pollination and fertilization and the result of this is that the fruit generally ripens very slowly and very variably.

M. Guignard observed that in *Vanilla aromatica* the ovary of the opened flower has an average measurement of 4 cm. in length, the placentas hardly showing the papillae which develop into ovules. If certain flowers are pollinated the pollen germinates at the end of several hours and the gynoeceum and the ovary open as early as the next day, the evidence of their influence. Eight days later the ovary has a length of 9 cm., the ovular nucellus is formed and the internal integument appears at its base in the form of an annulary papilla. The pollen tubes which consist of six fascicles disposed two by two against the ovary wall descend to the middle of the cavity. One month later the ovary measures 15 cm. in length, in the greater part of the ovules the embryo sac has formed its sexual apparatus, the pollen tubes have reached the base of the ovary and have commenced to creep to the surface of the placentas.

It is only a month and a half after pollination, sometimes a little sooner, that fertilization takes place. The little embryo is formed and fills the embryo sac. The ripe seed is no larger than the ovule was at the moment of fertilization. There are no external characteristics which indicate whether one has a fertile or sterile seed—that is an ovule deprived of an embryo.

In *Vanda tricolor pallens* fertilization does not take place until six months after pollination and this interval may reach 10 months in *Vanda suavis Rollissoni*. It is about four months in *Angraecum superbum*, *Sobralia macrantha* and *Dendrobium nobile*, as the author<sup>38</sup> established in a new memoir. He has shown further that the time which passed between pollination and fertilization of the ovules was two months for *Eria stellata* and *Phajus grandifolius*; three to four months in *Cypripedium*, five months in *Epidendrum ciliare*, and *Cymbidium sinense*, six months or more in *Vanda*, *Cattleya*, *Stanhopea*, *Coelogyne*, *Saccolabium*, *Aerides*, etc. In indigenous species the time is much shorter—

<sup>37</sup> Sur les effets de la pollinisation chez les orchidées. *Compt. Rend. Acad. Sci. Paris.* 1886.

<sup>38</sup> *Ann. Sci. Nat. Bot.* VII. 4: 202-240. 1886.

ten days in *Orchis Morio*; three weeks in *Orchis latifolia*, *Ophrys arachnites*, and *Epipactis rubra*.

For these same plants, Paul Maury<sup>39</sup> gave the following figures: two or three days in *Neottia ovata*; five or six in *Orchis latifolia* and other species of orchids; nine or ten days in *Loroglossum hircinum* and in *Ophrys*.

M. Dieuzeide,<sup>40</sup> as well as Darwin, maintains to have established that self-fertilization is the rule in *Ophrys apifera*, a plant which is very abundant and very fertile in favorable soil. *Ophrys muscifera* and *O. aranifera* gives seed only when cross-pollinated.

### PARTHENOCARPY IN ORCHIDS

In the conclusion of his Memoir, Guignard stated that everything indicates that the ovules are subordinate in development to the ovary. In exotic orchids, the thickness and length of this organ are often very pronounced before the appearance of ovules. Gaertner<sup>41</sup> devotes one special chapter to the thickening of the ovary without preliminary pollination, from which there results a pseudo-fruit. Among the plants observed he cited the orchids. If a closer study is made of the causes of this growth and if the physiological aspect of the question is considered one is led to admit, he states, that if the formation of pollen tubes is necessary for the growth of the ovary and its contents they do not have a specific effect on the latter. The pollen tubes borrowing from neighboring tissues substances which they need for their growth bring about an afflux of nutritive material. Anything capable of acting thus should produce the same result. Do we not have examples of analogous facts in various cases of parasitism, where the cells soon become hypertrophied and then multiply plentifully like those which are invaded by *Synchytrium*, or again as in the formation of galls developed under the influence of a foreign vegetable or animal organism? If the phenomena are different morphologically, physiologically they are comparable. Well established facts have shown that parasites may produce the same effects as the pollen tubes in inducing a growth of ovaries and placentas and a development of ovules.

### OBSERVATIONS ON THE STERILITY OF EXOTIC ORCHIDS

I owe to Doctor Jean Gratiot, vice president of the Committee on Orchids of the National Horticultural Society of France, the following personal observations on total or partial sterility of exotic orchids.

The pollination of exotic orchids always done artificially in our greenhouses resulted in: First, the production of fruit containing seeds with good embryos; second, fruit whose seeds were partially or totally sterile; third, absolutely empty fruit, and fourth, absence of fruit.

Total or partial sterility may result when there is pollination between two plants of the same genus and species, or when pollination is between two

<sup>39</sup> Sur la pollinisation des Orchidées indigènes. Comp. Rend. Acad. Sci. Aug. 2, 1886.

<sup>40</sup> Proc.-verb. Soc. Linn. Bordeaux. 1922: 40.

<sup>41</sup> Gaertner. Versuche und Beobachtungen ueber die Befruchtungsorgane der vollkommeneren Gewächse. Stuttgart. 1844.

plants which are widely separated botanically. In the first case, sterility may be due to general causes which were present in all the plants.

The pollinations may be neatly made aseptically to avoid carriage with the pollinia to the stigma of fungus spores or any other germs which would infest the gynostemium and destroy the ovules, as happens frequently with preserved pollinia. Molds can develop on the pollinia themselves and destroy them partially or totally. The pollen masses which had been wet nearly always lost their fertilizing power. Lastly, whatever care may have been exercised in their preservation they become unfit for pollination after a certain length of time which varied according to the genera and species. Pollen of *Phalaenopsis* and *Vanda* preserve their power of fertilization a long period while the viability of pollen of *Cypripedium* is of very short duration.

Many causes of sterility are general and exist in the pollination of plants which are closely related botanically as well as in those of more or less distant parentage. A cause of sterility that concerns plants which are botanically different rests in the disproportion in length and breadth of pistils, and above all of ovaries. The success of pollinations is much more assured if the organs are well proportioned.

In the hybrids from hybrids of the third, fourth, fifth or sixth generations or more it seems that the plants end in becoming to a greater or lesser degree sterile from poor constitution of pollinia or of ovules.

It is fitting to repeat, meanwhile, that sterility is nearly always the rule when it is a matter of fertilization between plants botanically distant. But one may say also that certain species of orchids which seem to be closely related nearly always give only negative results when one tries to associate them. It is difficult for example to cross *Phalaenopsis Luddemanniana* by *Phalaenopsis amabilis*, while the reciprocal fertilization has much less chance of remaining sterile. The cause of this sterility remains unknown. Elsewhere even though the pollinations were made under conditions which seemed to guarantee success, fruits with nearly perfect embryos were obtained while others contained only a variable and at times very reduced proportion without any apparent reason.

The external appearance of a fruit is deceptive and does not give any indication of the quality of its contents; nevertheless, a well constituted fruit which is normally developed offers more assurance than another poorly formed which had a prolonged growth. In every case one should resort to the microscope to be assured whether or not the seeds contain embryos.

The questions which I have reviewed are still not cleared up. It is only by multiplying observations and experiments that we may perhaps succeed in clarifying them.





## ABORTIVE AND STERILE APPLE POLLEN

ERLING KVAALE

*Sogndal i Sogn, Norway*

For most of the workers on sterility and fertility in apples it is a known fact that there exist quite a few varieties which yield pollen of low viability as far as it can be determined by germinating tests in artificial media. Tests are made in different countries under somewhat varied climatic conditions, but the results seem to be as nearly identical as they can be. Even at the same place tests will vary to some extent. Different culture media and changes in temperature will influence the results. But everywhere, as far as I have seen the results, the varieties with high germination will show this, and the ones with low germination will retain their low viability under all conditions. The question then arises to what extent will this low germination, in the natural performance of germination and fertilization in the flowers, influence the setting of fruits—which question in this connection is most important.

Will the same percentage that germinates in artificial media in cases of low viability be able to germinate and to fertilize ovules, and thus induce setting of fruit? Or, will the weakness which follows the low germination also be the same after germination on the stigma. And another question is: can there be fruit setting without fertilization, and to what extent?

We know that varieties such as Ribston and Gravenstein under certain conditions can give well-sized and mature fruits without seeds, and Cellini even more so; but as a rule under conditions which we want to have in the orchard, seeds in the apples are the natural thing.

In each pollination there is a considerable number of pollen grains applied on each stigma, so many that even a small percentage of good pollen would be enough for fertilization. On the other hand, we know that an abundance of good pollen applied to the stigma fails to set fruit in many cases, which indicates that there are many factors that may influence the setting of fruit. Some of them may be accounted for but some of them not.

The following discussion will deal only with the germination of pollen and the setting of fruit when pollen of low germination is used. My own work on this line was started in 1921, but did not get full attention before 1922, when most of the germinating tests were made. The crossings presented in tabular form are the results from 1923 and 1924 combined, although there would be reason to present them separately if the question was about technical points in the crossing work. For in 1923 all isolations were in paper bags, while in 1924 houses were built around trees or part of them. But the result after the two years are so uniform that the technique employed seems not to have influenced the results.

The selected list of crossings done by other workers, who used the pollen

from varieties with low germination, contains all crossings in which the particular varieties in question were used.

### THE POLLEN OF APPLES

*Morphology.* An examination of pollen grains of apples under the microscope shows in most cases two distinct groups of pollen, viz.: one class of small, narrow, colorless and empty grains, and the other with larger and colored grains filled with cytoplasm and somewhat different as to size and form.

The exact proportion of empty grains in different varieties can be determined by adequate countings. The following list will show the approximate number for a few varieties:

Astrachan red .....	5%	McIntosh .....	40%
Astrachan white .....	5%	Nansen .....	50%
Baldwin .....	25%	Red Stettiner .....	75%
Cox's Orange .....	50%	R. I. Greening .....	50%
Fuhr .....	1%	Ribston .....	10%
Gravenstein .....	50%	Roter Eisenapfel .....	60%
Harberts Reinette .....	50%	Aker .....	40%
Kaupanger .....	2%		

Of what significance these numbers of empty pollen grains may be we can not say at present. But this list compared with the germination will show some connection between a low germinating power and a high percentage of empty grains, although, as in the case with Ribston, there is low germination and a low percentage of empty grains, and in McIntosh a relatively good germination and about 40 per cent empty grains. MacDaniels (10) found two-thirds of the McIntosh pollen to be empty grains, and a high percentage also for Baldwin and Rhode Island Greening.

That we have to deal with results of irregularities in the division of the pollen mother cells seems very likely.

As to the size of pollen filled with cytoplasm I have not found so much difference. The pollen in dry condition as they are when the anthers open seem not to have spherical form, but more or less pointed at two ends and probably somewhat flattened. As soon as they are in water or another fluid, they look spherical or triangular. A medium size for pollen grains of Wealthy, McIntosh, Red Astrachan and Red Stettiner seems to be 20-25 by 40-55 microns. To arrange any groups of the pollen according to size, making more than the two groups, viz.: (1) empty and (2) filled grains, seems problematical. But a distinction (apparently after the germination tests) has been made by Beaumont and Knight (4) between three groups that did not germinate. Shoemaker (13) gives the measure of 1,000 grains for each of the varieties Delicious, Paragon and Stayman. The pollen grains of Delicious have a range in size from 20 to 55 microns, and most of them are around 45 to 50; Paragon has a range from 20 to 60 microns, and Stayman from 15 to 60 microns with the highest number around 45 and 50.

One instance which seems to be unique among apple varieties is the sticky nature of Winesap pollen. This variety is not grown in our country, but when I was at the Geneva Station, I made tests with this pollen too, and

found that the grains stick together in lumps. The way it looked, the pollen from half of the anther seemed to be in one lump. The germination apparently was good.

*Physiology of Apple Pollen.* The first work done on the physiology of apple pollen which is of interest in this connection was by Booth.\* He seems to have found the same difference in germination of pollen as is found in this work. But in one way or another the result of his work was not paid much attention to, neither was it taken as guide for further crossing, nor was it published.

Ewert (5) did some work on pollen, but it is of less interest when the question is about differences between varieties. The same is the case with Adams (1).

By far the most complete work on this line was done by Florin (6). The results of his work are so close to what is reported here, that a discussion of it does not seem necessary. Beaumont and Knight (4) who tested 17 varieties found a range in germination from 28.3 per cent and up to 89.1 per cent. As the lowest germination was in Gilbert Winesap (Paragon) and Stayman, it shows the same as other tests of this kind but the percentage for the two named varieties is somewhat higher than what is reported in other works; and the same is the case for the length of the pollen tubes. MacDaniels (10) found for Baldwin, Rhode Island Greening and Arkansas even lower percentage than here reported, and for McIntosh not more than 24 per cent. For Delicious and Cortland it is about the same as in this report.

Kobel (9) found in Switzerland that Gravenstein, Jacques Lebel, Schoner von Boskoop, Winterzitronenapfel Menznauer Jaegerapfel, Roter Eisenapfel, Rheinischer Bohnapfel, Staefner Rosenapfel and Baldwin had an average lower than 20 per cent of germination.

*Tests of germination.* As already mentioned, most of the germination work was done 1922. Additional work was done at the New York State Agricultural Experiment Station, 1925. Also some tests were done again in this country 1926, in order to be sure if there might be any differences between different years and in different countries. But in all cases, as far as the work was done on the same varieties, no differences was found more than what can be observed by making several tests of the same variety at the same time.

When the work started three varieties of apples (besides other fruits) were tried in seven different media. As the medium made up by 0.5 per cent agar and 5 per cent glucose in water showed about the best results for a good growth of the pollen tube, this was used for all later tests, with the exception of the tests done at Geneva, where 5, 10 and 15 per cent glucose and 0.5 per cent agar was used. In these tests 10 and 15 glucose gave the best results. Cane sugar in water alone, and an addition of different percentage of gelatine gave short pollen tubes which burst at the tips. But the percentage of germination was about the same as for tests in agar-media.

---

\*Booth was at the New York State Agricultural Experiment Station, and the work in question was done some time after 1900.

Further, in the following table the pollen-tube length given for the varieties with low germination is for the longest tubes, and only a few of them were as long as the measure given, while the length given for the rest of them will be about the average in each culture.

TABLE 1  
GERMINATION OF APPLE POLLEN

Variety	Number of pollen grains	Percentage of germination	Length of pollen tubes in microns
Aga .....	95	90	760
Alexander .....	110	66.4	822
Annie Eliazbeth .....	54	76	1748
Antonovka .....	79	81	1298
*Arkansas .....	666	11.4	....
Arreskov .....	107	90.5	....
Astrachan red .....	94	70	1473
Astrachan white .....	113	62.8	....
†Baldwin .....	798	12.3	....
*Banks .....	985	10	....
Bergvikseple .....	88	84	1680
Bramley Seedling .....	86	20.9	662
Brureple .....	87	77	1544
Casseler Reinette .....	63	88.9	1645
†Charlamovsky (Oldenburg) .....	94	87.3	2437
Cellini .....	126	88.1	2407
*Cortland .....	440	70.2	....
Cox's Orange .....	75	69.3	1948
Cox's Pomona .....	65	98	1663
Crasnyi letnyi .....	81	80	2138
*Delicious .....	536	89.6	....
Devonshire Quarrenden .....	79	97.5	841
Dronning Louise av Danmark .....	120	79.2	679
Early Harvest .....	62	79	735
Early Rivers .....	169	79.9	536
Early Rivers Peach .....	97	87.6	602
Edholm .....	49	75.5	678
Ekely .....	74	91.9	2217
*Esopus Spitzenburg .....	91	66	....
Fayry Queen .....	27	96.3	2185
Frogmore Prolific .....	63	89.5	1358
Fuhr .....	104	83.6	2027
Garfield .....	126	88.1	1362
Gooseberry .....	76	56.6	540
Granat .....	134	96.3	2459
†Gravenstein .....	238	13	810
Hampus .....	78	46.2	2261
Hans Matthiesen .....	64	84.4	1755
Harberts Reinette .....	153	18.9	722
Haugmanns .....	71	71.8	1140
Italiensk Muskat .....	119	78.2	2218
Kaupanger .....	184	2.8	....
Keswick Codlin .....	85	96.5	1553
Klareple .....	100	75	519
Kjolstad .....	121	20	557
Lane's Prince Albert .....	99	57.6	447
Langtons Nonsuch .....	63	98.4	2058
Lindemanns Pigeon .....	87	97.7	1140
Magnus .....	178	19.7	865
†McIntosh .....	102	56.9	1077
McMahon .....	90	84.4	1140

\* Tried at the Geneva station.

† Tried both in this country and at the Geneva station.



TABLE 1—Continued

Variety	Number of pollen grains	Percentage of germination	Length of pollen tubes in microns
Nonnetitte .....	96	55.2	796
Newton Wonder .....	48	52.1	1693
Okabena .....	63	95.2	1488
Peasgood Nonsuch .....	98	58.1	1995
Pewaukee .....	73	83.6	1207
Plum Cider .....	91	84.6	1824
Porteple .....	168	88.1	2090
Prince Albert of Prussia.....	56	51.8	1315
Prinsesse Noble .....	111	71.2	900
Paronapple .....	112	31.3	1070
Red Reinette (Nansen) .....	149	4	108
Red Stettiner .....	240	6.3	900
Reinette Coux .....	112	75.9	1473
†R. I. Greening .....	114	28	1202
†Ribston .....	145	21.4	525
Ringstad .....	120	62.5	1223
Rodovitka .....	82	80.5	1021
*Rome Beauty .....	119	90.5	....
Rosenhager, Swedish .....	78	92.3	1169
Rosenstrips .....	143	94.4	1686
Roter Eisenapfel .....	162	19.8	1183
Scotts Winter .....	75	45.3	2014
Signe Tillisch .....	86	86	938
Soplime .....	95	92.6	1370
*Spy, Northern .....	114	90	....
*Stayman .....	631	6.5	....
Suinula Vaxim .....	43	67.4	741
Safstaholm .....	124	64.8	722
Sornes .....	205	69.8	1018
Torstein .....	147	90.5	1220
Transparente blanche .....	88	52.3	750
Transparente de Croncelles .....	73	60.3	1519
Vinterkronen .....	46	89.1	469
Virginsk Roseneple .....	103	55.3	1084
Warners King .....	67	14.8	230
†Wealthy .....	78	85.9	770
Williams Favorite .....	105	79	336
*Winesap .....	150	46.7	....
Wonder of Chelmsford .....	79	78.5	1772
Akero .....	102	42.2	418

\* Tried at the Geneva station.

† Tried both in this country and at the Geneva station.

We find in TABLE 1 that there is a range in germination from as low as 2.8 per cent in the case of Kaupanger, and up as high as 98.4 per cent for Langtons Nonsuch. Although most of the varieties rank above 50 per cent, there is a considerable number lower than that. Fifteen varieties are lower than 30 per cent. As some of these are to get special attention in the crossing work, they will need to be listed separately. They are, Arkansas, Baldwin, Bramley Seedling, Gravenstein, Harberts Reinette, Kaupanger, Kjlstad, Magnus, Nansen (Red Reinette), Red Stettiner, Rhode Island Greening, Ribston, Roter Eisenapfel, Stayman and Warners King. Banks must be regarded as identical with Garvenstein. Shoemaker (13) has found that the low germination in pollen is connected with irregularities in the division of the pollen mother cells. Paragon and Stayman are named as samples, while a variety as Delicious seems to have a more regular division. This corresponds

with my observations on Gravenstein and some other varieties, although I did not come to any conclusion in my work, because I thought that the irregularities might be due to bad fixation (preliminary work).

### THE CROSSINGS

The crossing work herein reported has been carried on in close connection with the work on pollen physiology. After some preliminary work in 1922 when only the number of bags were taken, the flowers have been counted, and later on in summer and fall the number of fruits was taken. In 1922, 1923 and 1926 paper bags were used for isolation of the flowers, while in 1924 houses were built around trees or part of them. The houses were built of glass, parchment paper (as top) and cheese cloth. As to the desirability of one of these methods in preference of the other, it is to say, that the houses seem to afford an ideal isolation and safe working facilities. But as the bags

TABLE 2  
RESULTS OF THE CROSSINGS DONE 1923 AND 1924 COMBINED

Seed parent	Pollen parent	Number of flowers	Fruits set
Cox's Pomona	× Ribston	66	0
Cox's Pomona	× Roter Eisenapfel	58	1
Cox's Pomona	× Nansen	32	0
Gravenstein	× Nansen	340	0
Gravenstein	× Red Stettiner	190	0
Gravenstein	× Harberts Reinette	65	0
Gravenstein	× Ribston	113	1
Filippa	× Gravenstein	22	2
Filippa	× Ribston	23	0
Fuhr	× Roter Eisenapfel	73	0
Fuhr	× Gravenstein	122	0
Fuhr	× Ribston	86	0
Nansen	× Gravenstein	25	0
Ribston	× Gravenstein	21	0
Rodovitka	× Gravenstein	75	10
Rosenstrips	× Gravenstein	76	0
Torstein	× Red Stettiner	41	0
Transparente blanche	× Gravenstein	32	0
Williams Favorite	× Gravenstein	15	0
Gravenstein	× Transparente blanche	113	6
Gravenstein	× Rodovitka	198	19
Gravenstein	× Akero	121	23
Gravenstein	× Fuhr	158	14
Gravenstein	× Bellefleur de France	56	1
Gravenstein	× Charlamovsky	315	23
Gravenstein	× Alexander	103	19
Gravenstein	× Rosenstrips	66	4
Gravenstein	× Safstaholm	105	32
Gravenstein	× Astrachan white	51	6
Cox's Pomona	× Alexander	60	5
Cox's Pomona	× Fuhr	41	1
Filippa	× Akero	25	9
Filippa	× Alexander	22	0
Fuhr	× Alexander	40	5
Ribston	× Akero	19	3
Rodovitka	× Rosenstrips	49	19
Rosenstrips	× Astrachan white	122	27
Torstein	× Akero	22	5
Torstein	× Alexander	23	3
Transparente blanche	× Akero	21	2
Transparente blanche	× Safstaholm	63	8

are much cheaper and seem to give about as uniform results as the houses do, it should be satisfactory to use bags.

The plan in the crossing work here reported has been to compare the group of varieties which is characterized by the lowest germination with some varieties of good germination. In all cases when only parts of the trees were used for the crosses, there was a good set of fruit on the rest of the trees. When the whole tree was isolated for the test, care was taken not to give any of the two groups preference as to location on the tree.

As will be seen from TABLE 2 the varieties are divided into two groups. The first group is made up by the ones that did not set fruits, or they set only few. All these pollen varieties belong to the group with less than 30 per cent germination.

In the second group are the successful crosses. It will be noted that only in one case was there no fruits at all, that is, in the cross Filippa  $\times$  Alexander, but there were not enough blossoms to show that this particular combination was incompatible. It also should be added, that in the cross Rodovitka  $\times$  Gravenstein which gave 10 per cent set, further tests are needed before it can be said that this is a case where Gravenstein pollen gives fruit. An error in the construction of the house made the conditions different from all others.

From these crosses as a whole it seems very likely that pollen with a germination as low as 30 per cent or lower will not be dependable for crossing with other varieties, at least not under conditions as they are in this country.

## RESULTS REPORTED BY OTHERS

Because the crosses here reported on do not cover all varieties with poor pollen, and the ones tried are not always in sufficient number to give a final proof for the cross-sterility which results from bad pollen, an extract of the results of some other works, seen in connection with what is reported here, will not be out of the way.

TABLE 3 is a selection of varieties which we know have a low percentage of germination and have been used as pollenizers in different parts of the world.

All the pollen varieties named by Kobel (9) have low germination, and he had a small number of flowers in each combination. But, as far as it goes, it gives uniform results, which strongly support the results obtained in this country. The work of Ballard (3) with Stayman and Gravenstein points exactly the same way. The percentage of setting of fruit after bad pollen is too low to be classified even as a medium set of fruit. Morris (11) got a good set of fruit from Baldwin, Gravenstein and R. I. Greening in a few cases, but usually they did not set any fruit at all. The only one who got fruits from most crossings with Baldwin and R. I. Greening was Sax (12). While the percentage usually is low, only one cross (R. I. Greening  $\times$  Baldwin) failed entirely. Sax worked at the same place as did Gowan (8); but the latter obtained fruit only in two cases out of ten when Baldwin and R. I. Greening were used as pollen varieties. The few clusters used, however, may

be insufficient for a trial of this kind. But also the report from Gourley (7) points the same way, although he does not say how many flowers he used for the experiment.

TABLE 3  
CROSSINGS MADE BY OTHERS

	Number of blossoms	Number of fruits	%
Kobel (9)			
Schoner von Boskoop × Winter zitronenapfel .....	16	0	..
Schoner von Boskoop × Stafner Rosenapfel .....	26	1	4
Schoner von Boskoop × Bohnapfel .....	21	0	..
Winterzitronen × Schoner von Boskoop .....	27	0	..
Winterzitronen × Stafner Rosenapfel .....	25	0	..
Winterzitronen × Bohnapfel .....	23	0	..
Gravenstein × Witerzitronen .....	23	0	..
Gavenstein × Stafner Rosenapfel .....	22	0	..
Gravenstein × Bohnapfel .....	19	0	..
Bohnapfel × Schoner von Boskoop .....	16	2	12
Ballard (3)			
Early Ripe × Stayman .....	285	8	2.8
Grimes × Stayman .....	878	41	4.6
Ingram × Stayman .....	242	0	..
Mother × Stayman .....	606	4	0.6
Nickajack × Stayman .....	371	0	..
Rome × Stayman .....	604	0	..
Stayman × Gravenstein .....	300	0	..
Williams × Stayman .....	14	0	..
Yellow Transparent × Stayman .....	212	0	..
Morris (11)			
Chenango × Gravenstein .....	93		3.25
Esopus × Stayman .....	108		..
Fallawater × R. I. Greening .....	25		..
Jonathan × Baldwin .....	28		..
Jonathan × Gravenstein .....	74		..
Jonathan × R. I. Greening .....	64		..
Jonathan × Stayman .....	85		..
Longfield × Baldwin .....	26		25.9
Longfield × R. I. Greening .....	25		44
McIntosh × Baldwin .....	26		38.46
McIntosh × Stayman .....	168		..
Rome × Stayman .....	77		..
Twenty Ounce × Gravenstein .....	38		..
Wagener × Gravenstein .....	65		20
Wagener × R. I. Greening .....	47		6.9
Wagener × Stayman .....	99		..
Sax (12)			
Ben Davis × Baldwin .....	515		2.5
Ben Davis × R. I. Greening .....	637		2.5
Baldwin × R. I. Greening .....	342		3.5
Russett × Baldwin .....	666		4
Russett × R. I. Greening .....	347		22
R. I. Greening × Baldwin .....	375		..
Spy × Baldwin .....	232		2
Spy × R. I. Greening .....	219		2
McIntosh × Baldwin .....	179		13
McIntosh × R. I. Greening .....	255		8
Gowen (8)			
Ben Davis × Baldwin .....	11	1	
Dutchess × Baldwin .....	20	0	
Golden Russett × Baldwin .....	2	0	
Spy × Baldwin .....	12	1	
Red Astrachan × Baldwin .....	4	0	
R. I. Greening × Baldwin .....	2	0	
Baldwin × R. I. Greening .....	2	0	
Ben Davis × R. I. Greening .....	2	0	
Golden Russett × R. I. Greening .....	2	0	



TABLE 3—Continued

	Number of blossoms	Number of fruits	%
Spy X R. I. Greening .....	2	0	
Gourley (7)			
Grimes Golden X Baldwin .....			
Wealthy X Baldwin .....			2.2
Rome X Baldwin .....			1
Spy X Baldwin .....			..
Nero X Baldwin .....			..
Stayman X Baldwin .....			..
McIntosh X Baldwin .....			1.3
Jonathan X Baldwin .....			..

As a whole, this selection of crosses from different workers shows, in spite of the few exceptions, the same results as I have got in this country. It is to be noted, however, that Auchter (2) in his work found incompatible varieties among the ones with good pollen as well as among those with bad pollen.

### CONCLUSION

Germination tests with pollen from most of our cultivated varieties of apples show that there is a group of varieties with pollen of low viability, the percentage of germination being lower than 30, while the greatest number of varieties yield pollen with high germinating power, usually between 50 and 100 per cent.

Tests as made in different countries, show no greater differences for any one variety than are to be observed by making different tests at the same place. The percentage of germination also seems to be much the same for the different years, so that the percentage of germination is a characteristic for each variety.

The crossing work done in this country and other places seems to indicate that the varieties which have pollen of low germination are not reliable pollenizers for common orchard practice, although there may be some setting of fruit. In the cases where the work was laid out for the purpose of finding out about the bad pollen, a rather uniform result was obtained, while in the cases where pollen varieties were chosen without this in view, a less uniform result was obtained.

### ACKNOWLEDGMENT

The writer is indebted to Professor H. Misvaer of the Agricultural College of Norway, who opened the college orchard for my work, also to A/S Norsk Varekrigs Fond for its appropriation of a sum of money which made possible a comparison of methods of isolation of the flowers.

### LITERATURE CITED

1. Adams, J. On the germination of pollen grains of apples and other fruits. *Bot. Gaz.* 61: 131-147. 1916.
2. Auchter, E. C. Apple pollen and pollination studies in Maryland. *Proc. Amer. Soc. Hort. Sci.* 1921: 51-80.

3. Ballard, W. R. Methods and problems in pear and apple breeding. Maryland Agr. Exp. Sta. Bull. 196. 1916.
4. Beaumont, J. H., and Knight, L. I. Apple pollination germination studies. Proc. Amer. Soc. Hort. Sci. 1922: 151-163.
5. Ewert, Richard. Die Widerstandsfähigkeit der einzeln Organe der Obstblüte insonderheit des Blütenpollens gegen Frost. Zeitschr. Pflanzenkr. 20: 65-76. 1910.
6. Florin, R. Zur Kenntniss der Fertilität und partiellen Sterilität des Pollens bei Apfel- und Birnensorten. Acta Horti Bergiani 7: 1-39. 1923.
7. Gourley, J. H. The problem of the unfruitful tree. Fifty-sixth Ann. Rep. Ontario Fruitgrowers Assoc. 1924.
8. Gowen, John W. Self-sterility and cross-sterility in the apple. Maine Agr. Exp. Sta. Bull. 287.
9. Köbel, F. Die keimfähigkeit des Pollens einiger wichtiger Apfel-und Birnensorten und die Frage der gegenseitigen Befruchtungsfähigkeit dieser Sorten. Landw. Jahrb. d. Schweiz 38: 461. 1924.
10. MacDaniels, L. H. Pollination studies with certain New York apple varieties. Proc. Amer. Soc. Hort. Sci. 1925: 87-96. 1926.
11. Morris, O. M. Studies in apple pollination. Wash. Agr. Exp. Sta. Bull. 163. 1921.
12. Sax, Karl. Sterility relationship in Maine apple varieties. Maine Agr. Exp. Sta. Ann. Rep. 1922.
13. Shoemaker, J. S. Pollen development in the apple with special reference to chromosome behavior. Bot. Gaz. 81: 148-172. 1926.











581.2 I61



a39001



007148771b

581.2  
I61

581 2 I61

INTERNATIONAL CONFERENCE ON FLOWER AN

INSERT BOOK  
MASTER CARD  
FACE UP IN  
FRONT SLOT  
OF S.R. PUNCH

MASTER CARD

6LOBE 90144-0



UNIVERSITY OF ARIZONA  
LIBRARY



